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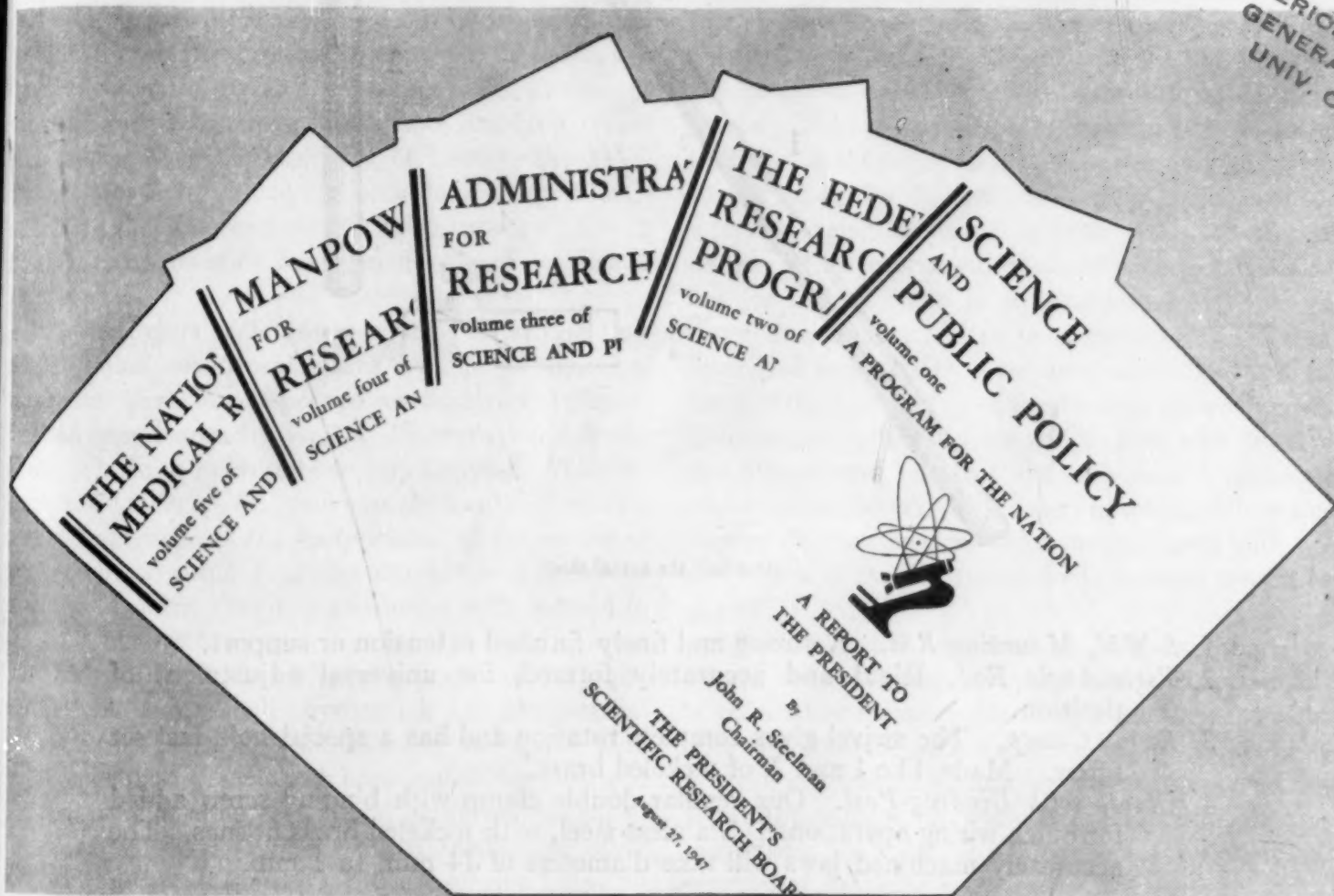
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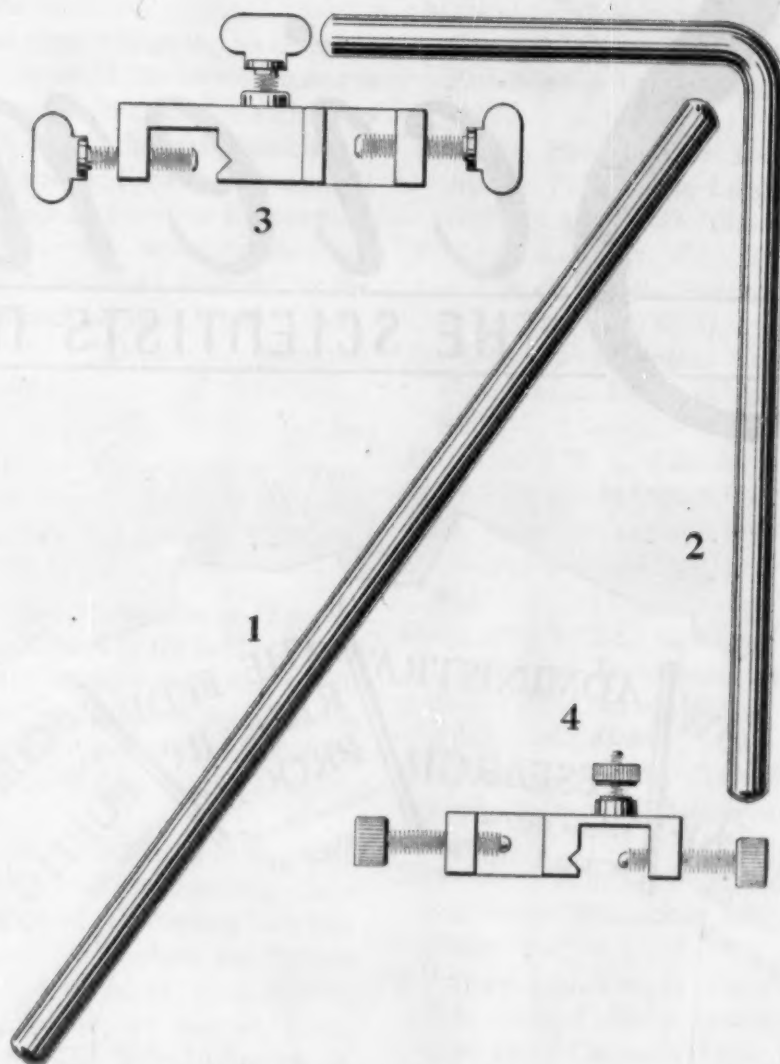


Among the various special sessions on the program of the AAAS Meeting, now being held in Chicago, is one on "Science in National Affairs," in which the President's Scientific Research Board Reports will be discussed. Federal Research Programs in the Civilian Agencies and in the Armed Services are to be discussed, respectively, by E. U. Condon and L. R. Hafstad; Administration of Federal Research, by W. V. Lambert; and the National Medical Research Program, by D. W. Bronk. J. Donald Kingsley will preside.

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Index Issue

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The Chemical Components of Onion Vapors Responsible for Wound-healing Qualities

Edward F. Kohman

Campbell Soup Company, Camden, New Jersey

THERE IS PROBABLY NO OTHER FOOD TO which is attached so much legendary history as is the case with the onion, although Pliny, the Naturalist, credits it with being the remedy for only 28 maladies—a moderate figure compared with the 87 for cabbage. Many of the legends have a bearing on physical health. Considering those dealing with evil spirits, the onion is involved even in mental health.

It requires but little observation to be aware of some strikingly peculiar chemical components in the onion. Their effect on the organs of the senses of smell and sight soon make this apparent. While man has been crying over onions since the beginning of history, like Mark Twain's weather, nothing has been done about it. It is surprising that the chemistry of onion vapors with such striking characteristics has been developed to such a slight degree.

In recent years facts have appeared indicating that certain onion constituents have notable physiological properties. That onion vapors possess bactericidal properties has been shown by Walker, Lindegren, and Bachmann (6). In a later publication, Ingersoll, Vollrath, Scott, and Lindegren (2) "provisionally identified acrolein or crotonaldehyde as the bactericides" in the vapors of both onion and garlic. From the data herewith presented it will be evident that this conclusion with respect to onions is incorrect.

In Russia, B. Tokin (4), over a period of 14 years, studied the bactericidal, phytoncidal, and protistocidal properties of over 150 plants. Onions and garlic were found to be most potent, with horse-radish, peppers, and radishes next in order. Onion and garlic were lethal to white staphylococci, typhus, and a number of other bacteria, and to all protozoa, the latter being killed by exposure to onion vapor for 1–3 minutes. Tokin found that the onion vapors killed protozoa even more promptly than bacteria and termed the vapors "phytoncides." He also found that by chewing raw onion 3–8 minutes, and sometimes for only 1 minute, the buccal lining usually became completely sterile. Tokin judged the onion phytoncides to be extremely volatile because onion paste ceased to give off bactericidal vapors after 10–15 minutes. As will be evident below, instability rather than volatility is the true explanation.

Presented at the Meeting-in-Miniature of the Philadelphia Section, American Chemical Society, June 1947.

Recently, Lucas and Hamner (3) demonstrated that the sodium salt of 2,4-dichlorophenoxyacetic acid became a much more potent herbicide when dissolved in onion juice diluted with 20–30 times as much water as when it was dissolved in water alone.

Inspired by such properties and impressed with the idea that ancient and traditional remedies should not be regarded as naive and absurd, Toroptsev and Filatova (5), of the Tomsk State University and the All-Union Institute of Experimental Medicine of the USSR, studied the effect of onion vapors on purulent inflammatory wounds. They placed the paste of one or two onions in a shallow dish the size of the wound and exposed the wound to the vapors for 10 minutes, usually in two 5-minute intervals, using the paste of fresh onion for each exposure. These investigations started with 25 patients, but for lack of onion (a striking commentary on war times) the number had to be reduced to 11. Of these, seven had amputations of the arm, one of the thigh, and three of the foot. In two of the patients the wounds were complicated with gangrene and in one with frostbite; the others were purulent and contained streptococci, white staphylococci, and other bacteria. All wounds showed distinct purulent inflammation, some with odor and edema of the soft tissue. Some patients complained of pain.

After the initial treatment all wounds became rose colored instead of gray, and there was no more complaint of pain. After the second treatment purulence subsided, odor disappeared, and regeneration was induced. Regeneration began to lag in a few cases after several days treatment, suggesting possible overexposure. Without minimizing the value of synthetic preparations to the practice of medicine and surgery, the Russian investigators take the position that the so-called phytoncides also have their place. The following study suggests to a chemist, however, that not the onion but the synthetic laboratory should be their source.

Onions were ground expeditiously in 800-gram quantities through a household meat and vegetable grinder and introduced into a 2-liter, round-bottomed flask with short neck. This was fitted with a Kjeldahl connecting bulb, which in turn was attached by a short, heavy rubber tubing to a flask imbedded in a salt-water-ice mixture to serve as a condensing receptacle. The bore of the tubing must be large in order not to impede distillation. After the whole system was pumped devoid of air by a

"high-vac" pump, the flask containing the onion was alternately submerged in water at 50° C. and lifted out to be vigorously shaken. This alternate shaking and periodic immersion is necessary. Unless it is properly performed, the distillate will contain the constituents to be described in far-reduced quantities. The idea is to introduce heat into the mass of onion as fast as the evaporation of the distillate takes it out and to avoid heating any part of the onion mass appreciably above room temperature, since by overheating the ingredients of the distillate sought are decomposed. It required about 6 minutes to grind the onion, introduce it into the flask, and evacuate the system, and 12 minutes more to obtain an amount of distillate equal to 15 per cent of the weight of the onions.

The distillate was water clear, nitrogen free, and contained the lachrymal principle of the onion, as a tiny drop in the eye at once demonstrated unmistakably. There was only mild lachrymating effect if the eye was held against the mouth of the flask containing the distillate. This indicates that not the volatility of the lachrymal principle but the turgidity and succulence of the onion tissue, causing invisible droplets of onion juice to be dispersed, is the cause of man's weeping over onions. Whereas spooning a grapefruit may be said to deliver a macro squirt on one's tie, cutting an onion delivers a micro squirt in one's eye. The distillate had an onion odor but contained none of the essential oil to which the flavor of the onion is due; the residue retained its full onion flavor. In a short time the water-clear distillate began to take on a milky appearance and, if held until the succeeding day, the density of the milkiness was comparable to that of skimmed milk. By that time it had, however, lost every vestige of lachrymal effect or any irritation in the eye even though the distillate was stored in a closed container to prevent any escape.

Acid hastened the development of the milkiness; alkali inhibited it. Bromine almost instantaneously clears up the milkiness and produces sulfate. But if bromine is added promptly to the distillate before any milkiness develops, practically no sulfate is formed, even with heat and long standing in the presence of bromine, and the milkiness does not develop thereafter. If, after the milkiness fully develops, the distillate is again distilled under the same vacuum and temperature, the milkiness remains as a gummy residue for which no satisfactory organic solvent has been found, although bromine water momentarily clears it up with the formation of the usual quantity of sulfate.

In a typical experiment the clear distillate of five 800-gram batches of onion was collected in a tared flask from which it could be redistilled. After holding until polymerization was complete and redistilling under vacuum at low temperature there remained a residue weighing .108 gram. Upon treatment with bromine, 0.3400 gram of barium sulfate was obtained. This indicates slightly over 43 per cent of sulfur in the residue, representing the

polymerized material. The sulfur content of thiopropionaldehyde and thioallylaldehyde is, respectively, 43 and 44.4 per cent.

Because of the unstability of this sulfur-bearing lachrymating substance it is difficult to estimate its total quantity in onions, since loss is going on from the moment the onions are ground and during the distillation period. To arrive at an approximation, one 800-gram batch of onions was ground and subjected to distillation at once while in the meantime enough onions for 5 additional batches were ground in the shortest time possible. From

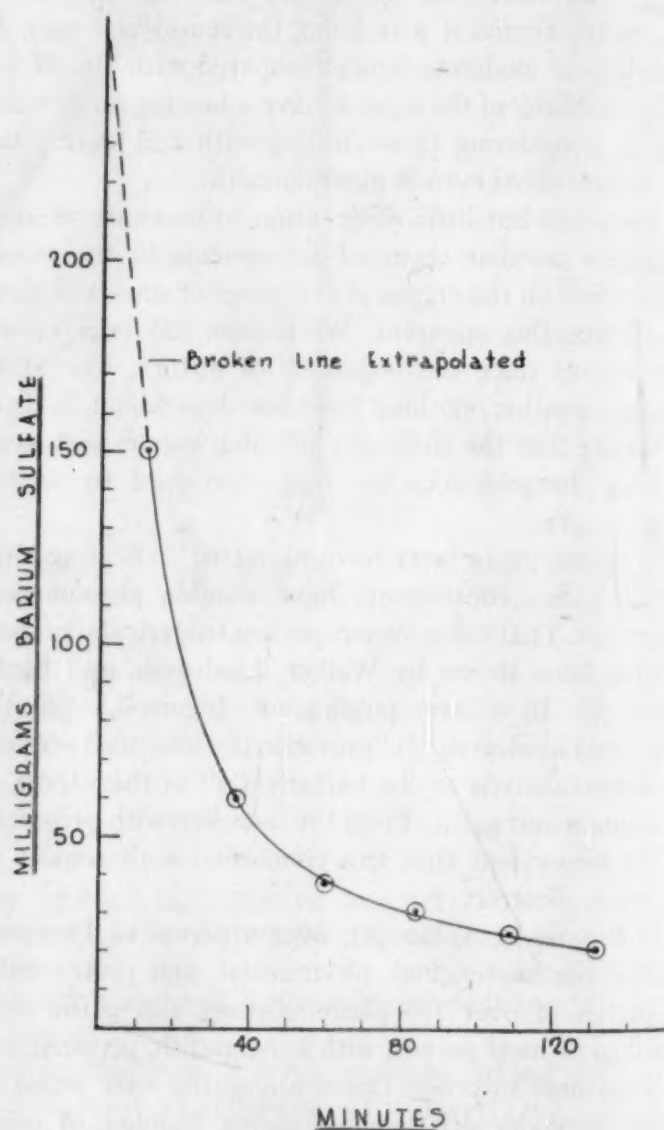


FIG. 1

these, at 24-minute intervals, an additional batch was subjected to distillation. By reckoning the time intervals from when the grinding in each case was half completed to when the distillation was half completed and plotting these against the barium sulfate obtained, Fig. 1 was constructed. By extrapolating this to zero time it is estimated that 800 grams of onion might yield 250 mg. of barium sulfate, which, calculated as thiopropionaldehyde, amounts to 100 mg./kilo. As indicated in the graph, prompt distillation yielded 150 mg. of barium sulfate. By using tubing with a larger bore between the flasks, 150 mg. were subsequently obtained.

These various reactions are interpreted to indicate that we are dealing with a thioaldehyde, the milkiness being the polymerized form. The fact that no sulfate is formed by bromine if added promptly after distillation and before polymerization is interpreted to indicate the reaction described by Douglass and Johnson (1), in which sulfonyl halide is formed from various thio-compounds. This meaningful reaction with bromine is the clue for future work toward the definite identification of this interesting thio-compound which apparently constitutes the lachrymal principle of the onion but which is not the essential oil that constitutes the onion flavor. It is suggested that it may be a step toward the synthesis by the onion of the essential oil. To our knowledge, no thioaldehyde has heretofore been demonstrated in nature. All are unstable, subject to ready polymerization.

From this low-temperature onion distillate obtained under vacuum the mononitro- and dinitro-phenyl hydrazones of propionaldehyde have been prepared and identified by melting points and analyses. The same quantity is obtained whether the preparation is made immediately after distilling, before any milkiness has developed, or after full milkiness has developed, or whether it is made from the clear distillate obtained by redistilling after full milkiness has developed, in which case the material causing the milkiness is left behind. If the phenylhydrazone preparation is made on the original clear distillate, a small amount of hydrogen sulfide is formed, which should theoretically occur if phenylhydrazine reacts with a thioaldehyde. The idea that thiopropionaldehyde is the lachrymal principle and that in the course of the development of the milkiness this is converted to propionaldehyde has not been disproved. But there seems no precedent for such a reaction. It seems more likely that the lachrymal principle is a sulfur compound present in addition to the propionaldehyde. The highly acid medium in which the nitrohydrazones are prepared would be expected to cause quick polymerization and thus prevent efficient reaction with the hydrazines.

In a typical experiment, the distillate from five 800-gram batches of onions amounted to 12 per cent of the weight of the onions and yielded 0.88 gram of dinitro-phenyl hydrazone after crystallizing twice from alcohol. This is equivalent to .21 gram of propionaldehyde or 52 mg./kilo of onion.

It is thus apparent that there is a sound basis for some of the traditions bearing on health which the onion has acquired. It is conceivable that eating raw onions has a curative effect on sore throat resulting from colds. There

is good reason for onion vapors having bactericidal and phytoncidal properties, and such vapors may be useful in healing wounds. But it is also apparent from the nature of the components of onion vapors that those compounds can be made available far more practically by synthetic chemical laboratories.

To control the fungus (*Urocystis cepulae*) responsible for smut in onions, formaldehyde is applied to the soil with onion seeds. The fact that this fungus no longer invades the seedling when it is several days old suggests that the seedling may protect itself by its own aldehyde. There is a tendency to ascribe any changes that take place in a broken or ruptured plant cell as being due to enzymic activity. But here we have a product removed from all contact with enzymes by distillation, yet undergoing profound chemical changes. It was recently announced that at Yale University 300 different chemical compounds have been obtained from the tuberculosis bacillus, which is a plant of but one cell. While the effect of enzymes is known to be profound, the imagination can easily picture many reactions between the numerous organic compounds that exist in organic materials such as food mixtures. The food chemist well knows that such reactions do occur spontaneously, unaided by enzymic catalysis, sometimes favorable, sometimes unfavorable. But he is still a long way from being able to control the media that contain such complex mixtures to direct the reactions according to his will.

Thus we have seen that propionaldehyde, which has been identified in the low-temperature vacuum distillate obtained from onions, is not the lachrymal principle in the onion. The lachrymal substance is concentrated in such a distillate, and the chemical reactions with it indicate a thioaldehyde. It is not so much the volatility of this substance that is responsible for the lachrymating effect of cutting an onion but the turgidity and succulence of the onion cell that disperses fine droplets of onion juice. Finally, if the components of onion vapors have a place in medicine and surgery, the organic chemist, not the onion, should supply them.

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The American Philosophical Society:

Abstracts of Papers Presented at Autumn General Meeting

The Discovery of an Oil Field

Paul D. Foote

Gulf Research and Development Company

Among the many smaller geologic basins distributed throughout the world, there are at least two gigantic basins in which great thicknesses of fine-grained marine sediments are interbedded with thick, porous, sandstone and limestone strata and are accordingly potential oil territory. One of these surrounds the present Gulf of Mexico and the Caribbean; the other borders the Persian Gulf. Although about half of the world's cumulative petroleum supply so far has been produced from the American basin, the Persian Gulf basin will undoubtedly prove to be the more productive area. The geophysical phase of a development that has led to the discovery of what appears to be the largest oil field in the world, now known as the "Burghan Anticline" in Kuwait, at the head of the Persian Gulf, is described. As a result of the pioneer work of the scientist, this general region has suddenly become one of the most important areas in the world from the standpoint of international politics and industrial development.

Benjamin Rush as a Promoter of Useful Knowledge

Lyman H. Butterfield

Princeton University

Benjamin Rush's devotion to projects for improving the conditions of human life sprang from his contact with ideas and leaders of the Enlightenment during his years abroad (1766-69). A particularly profound influence was exerted upon him by Benjamin Franklin, whom he met in London. Returning to America, Rush began those studies in a wide range of scientific and quasi-scientific fields of knowledge which he continued throughout his life and which are reflected in his voluminous correspondence.

The results of his investigations are embodied, in considerable part, in papers and addresses he read to the American Philosophical Society, of which he was for some 40 years an active member and officer. A representative paper is his letter to Jefferson on the need for, and means of, encouraging the production of maple sugar. Much more important is his oration on "the influence of physical causes upon the moral faculty," a statement basic to the then unborn science of psychiatry.

Rush's reputation as a medical scientist waned after his death and has not risen since. Though he served effectively as an advocate of experimental science, his own methods were uncritical, and his findings were warped by speculative preconceptions. His real contributions were in the marginal areas of medicine—to psychiatry, sanitation, veterinary science, and, above all, to medical education. His lifelong crusade for the popularization of medical knowledge, though little noted hitherto, may in the long run be considered his principal bequest to humanity. In disseminating the rules of good health,

hygiene, and diet, Rush was acting in accord both with the ideals of the Philosophical Society and his own vision of democratic America.

The Cultural Relations Attaché as an Instrument of International Understanding

Hayward Keniston

University of Michigan

The field of cultural relations as an aspect of American foreign relations is a recent development. Under the stimulus of the war, it has emerged as an instrument of national policy. There is a grave danger that its high purpose of promoting mutual understanding between peoples will be diverted into official propaganda in behalf of American political goals.

The years of experimentation, in Latin America and other countries, have established a pattern of the types of cultural interchange which are feasible and productive. They cover a wide range of individual and institutional relationships and have demonstrated the value of reciprocal exchange as a step toward common goals.

The Cultural Relations Attaché is the key figure in the development of the program. The activities which he must carry on determine the qualifications which he must possess for he must perform the dual function of representing to another people the intellectual ideals of the United States and of interpreting to the American people the cultural patterns of other lands. He must be supported by an adequate personnel to achieve the long-term goal of his office.

Reading and Visual Fatigue

Leonard Carmichael

Tufts College

The paper reports experiments on long periods of reading in an effort to study visual fatigue, conducted by Prof. Walter F. Dearborn, the author of this abstract, and a number of associates. A Kodachrome film shows the newly developed electrical recording technique used in the experiments. Twenty college and high school subjects read for two periods, each of which was 6 hours long. Half of these subjects read from the ordinary printed pages of a book; the other half, from a microfilm projection of book pages in a commercial microfilm projection device. Two books, one "dull" and the other "interesting," were read by each subject. Continuous recordings were made of every eye movement occurring during each of the two 6-hour periods for each subject. These records, totaling approximately 15 miles in length, were later edited and tabulated, and a number of indices, such as number of blinks, number of fixation pauses, number of regression movements, number of lines read, and the like, were studied. Careful evaluation of the results indicates that there was no statistically significant change in level of comprehension of the meaning of the material read or in any of the recorded characteristics of the eye movements during any 6-hour period. Some subjective

fatigue reports were given, but these were not related to any measurable loss of efficiency in reading. The conclusion is therefore presented that, for the typical group of subjects who were studied, continuous reading for this length of time produced no measurable fatigue. Some industrial, military, and educational implications that may be derived from the study are suggested.

Environment and Culture in the Last Deglaciation

Carl O. Sauer

University of California

Postglacial time differs from place to place, and the term is unsuited for dating the history of man. Critical markers of time may, however, perhaps be found in the culmination of the last glacial maximum and the ending of the subsequent deglaciation. For these, ages of 25,000 and 7,500 years, respectively, are suggested provisionally.

The pluvial climates that prevailed in low intermediate latitudes over areas now arid and semiarid, as in the American Southwest, are referred to a time anterior to deglaciation. During the latter the contemporary pattern of climates prevailed, with some exceptions in higher latitudes.

Deglaciation caused notable rise of sea level, the modern shore lines for the most part dating from the end of this period. It was also a major time of building of flood plains, of the formation of fresh and salt-water marshes, lakes, and lagoons. Optimum conditions resulted for plants and animals of shallow water, marsh, and alluvial habitats.

The Upper Paleolithic hunters gave way to the Mesolithic folk, with new basic economies. Among them were progressive groups of sedentary, riparian habits, developing skills in utilizing the resources of the waterside. The beginnings of cultivation and even of domestication fall into this period. The cradle of the new cultures is suggested as in southeastern Asia, from which the new ways of life spread also to the New World, by infiltration across high latitudes and then down the Atlantic seaboard.

Scholarship and Intelligence Problem

William L. Langer

Harvard University

An important feature of the recent reorganization of the armed services is the establishment of a new Central Intelligence Agency, which in the future will direct and coordinate all the intelligence activities of the Government in so far as they pertain to the national security. This involves not only the problem of collecting essential information, but also the work of research, analysis, and presentation—in other words, a largely scholarly function. The first serious efforts at exploiting the resources of American scholarship in the fields of the social sciences were made during the recent war, and the new agency is being organized on the basis of the experience then gained. There is no longer any question that, with the country's new position in world affairs, countless foreign problems require careful study by specially trained personnel. The new experiment promises well not only to meet an urgent governmental requirement, but also to react upon the work in the social sciences themselves, with particular reference to cooperation and integration. But an immediate and urgent

problem is that of finding suitable personnel, and this is a problem that should be viewed in the light of the larger program of government-supported research and training.

A Graphical Analysis of Respiratory Problems

Wallace O. Fenn

University of Rochester

A description is given of a diagram in which oxygen and carbon dioxide pressures in the alveolar air of the lung are plotted as coordinates. This diagram greatly simplifies the understanding of the mathematical relation between the rate of oxygen consumption, the rate of CO_2 output, the minute volume of the ventilation, and the varying composition of the inspired air. The advantages of the diagram depend upon the fact that it can represent by straight lines the following: (1) the CO_2 and O_2 tension breathing pure O_2 at any given altitude, (2) the various CO_2/O_2 exchange ratios (R.Q.) at any given altitude, (3) the volume of the alveolar ventilation per minute for a given rate of oxygen consumption and altitude or for a given rate of CO_2 output and altitude.

Not straight lines but curves can be drawn on the same chart representing (1) the percentage saturation of the arterial blood, (2) the cardiac output at given venous pO_2 tension, (3) the venous pO_2 at a given cardiac output and other quantities.

The use of this diagram for predicting the respiratory response is illustrated by experiments depicting the movements of the alveolar point while (1) rebreathing in a closed vessel in air or O_2 , (2) inhaling gas mixtures with high CO_2 in O_2 or air, (3) holding the breath in O_2 or air, (4) overventilating the lungs, (5) inhaling high O_2 or high N_2 mixtures, (6) ascending to high altitudes slowly or rapidly, (7) using pressure breathing to increase the pO_2 in the lungs and raise the altitude tolerance, and (8) breathing CO_2 mixtures at altitude.

The paper illustrates the great advance in our understanding of respiratory problems which came about during the war due to activities in many different laboratories.

A Fragment of a Greek Uncial Manuscript in Washington

Werner Jaeger

Harvard University

The Rare Book Department of the Library of Congress in Washington possesses an interesting single sheet of parchment which is of venerable age. It is covered with Greek text written in a beautiful, sloping, uncial hand. This parchment contains a fragment of early Christian literature, but there is no indication of either the title of the work or its author. The sheet is folded in the middle and thereby divided into two folia, both of which, recto and verso, are covered with text. These folia apparently represent the outer sheet of a quire which has at some time or other been removed from a manuscript of unusually fine quality. To what manuscript did this sheet originally belong, and is it lost or still in existence?

The present paper undertakes to identify the author and the work contained in the Washington fragment. It is possible, by comparing the handwriting on this sheet with that of other uncial manuscripts of the same type still extant, to identify the manuscript of which the fragment originally formed a part,

and to delimit the time when the separation occurred. The paper then proceeds to trace the history of this manuscript, on which the Washington fragment sheds interesting new light, through the Middle Ages and early Renaissance and tries to determine the period in which it was written and the part of the world from which it originated.

Ancient Mathematics and History of Civilization

O. E. Neugebauer

Brown University

In the history of the exact sciences the Hellenistic period will always be of special interest.

The analysis of the early development of mathematics reveals a variety of influences on this period, which extends from about the fourth century B.C. to the third century A.D. Least effective appears to have been Egyptian mathematics, because of its primitivity. The Babylonian contribution, however, was very great for the development of algebra as well as mathematical astronomy. Consequently, very different strata of "Greek" mathematics can be distinguished. Though classical "Euclidean" mathematics constitutes an enormous progress both in methods and results, its practical limits were soon reached. The survival of mathematical knowledge during the Middle Ages is to a large extent due to the continuity of the oriental tradition and to mathematical astronomy. It is especially the Persian and Arabic scientists who must be credited with the integration of the different layers of mathematical sciences into a larger unity sufficiently broad to provide the material needed for the development of mathematical astronomy, optics, and mechanics in the Renaissance.

Man's Greatest Illusion

Robert L. Schuyler

Columbia University

During almost the entire course of man's existence he has taken it for granted that he occupied a central position in the physical universe and that nonhuman objects and beings possessed attributes similar to his own. This illusion of anthropocentrism and anthropomorphism has been the greatest of all his illusions, and the most pregnant in its consequences.

We can no longer believe that we are literally at the center of the universe, as was believed by everybody down to about 10 generations ago; but it is still possible for us to believe that we are at the apex of creation, though there is no satisfactory basis for this belief, which appears to be nothing more than an instance of wishful thinking and a delusion of grandeur.

The remedy for it is the spirit of humility. Morally wise men of science and philosophy have recognized the necessity of humility for the acquisition of knowledge and the attainment of serenity; and in the realm of historiography the cure for the illusion that exalts our own age at the expense of all others is that form of humility known as historical-mindedness.

The Migration of the Methyl Group in the Body

Vincent du Vigneaud

Cornell University Medical College

The paper presents the experimental background which led the author and his co-workers to the concept that the body

is incapable of generating methyl groups for the methylation of certain nitrogen- and sulfur-containing compounds of the body, but is dependent on the presence in the diet of methyl groups in a utilizable form such as in methionine and choline and that a process of transmethylation is involved in certain steps in the intermediary metabolism of the body.

Direct proof of the transfer of the methyl group was afforded by tracing the migration of the methyl group by labeling it with deuterium. More recent work on transmethylation in which the methyl group has been labeled with C^{14} , the radioactive isotope of carbon, is also presented.

Streptomycin in the Treatment of Infections

Chester S. Keefer

Massachusetts Memorial Hospitals

and

Boston University School of Medicine

Streptomycin has been used extensively for the treatment of many infections that fail to respond to other chemotherapeutic agents. The results of its use in tuberculosis, tularemia, *H. influenzae* infections, brucellosis, and gram-negative bacillary infections are discussed. The discussion includes the over-all results in 3,000 cases.

Franklin-Greene Correspondence

William G. Roelker

Rhode Island Historical Society

An unpublished translation, in Franklin's unmistakable hand, of a Spanish letter to Miss Catharine Ray from Don Laureano Donado del Castillo, Cadiz Bay, June 7, 1751, is one of the rare items recently acquired by the American Philosophical Society. The original Spanish is awkward and formal, but the translation is in Franklin's charmingly fluent prose. Other letters added to the Society's library include the "Be gone Business" and the famous "multiplication" letter. These letters have always been in the possession of the family.

The correspondence between Franklin and Mrs. Catharine Ray Greene is the record of their long friendship, which endured from the day they met in 1754 until his death in 1790. Yet all these years they saw each other on but five occasions. Much has been written about it, but these letters reveal a gay, tender, "cheerful and agreeable Benjamin Franklin," who has never before been so disclosed to the public.

Catherine Ray, later Mrs. William Greene, had met Franklin at her sister Judith Hubbard's in Boston while on one of her periodic escapes from Block Island, 12 miles out at sea from Point Judith, the tiny islet where she was born and raised. "Katy," as Franklin addressed her, or "Catty," as she signed herself, was the daughter of Simon Ray and his second wife Deborah Greene, a great granddaughter of Roger Williams. Other members of the extensive Greene family to appear prominently are Nathanael, the Revolutionary General, who married Catty's niece, "Kitty" Littlefield, and his brother Elihu, who married Franklin's grandniece, Jane Flagg.

On the Franklin side, after Franklin himself, the most important is Jane, his favorite sister and wife of Edward Mecom of Boston. Jane early became attached to Catty, whom she had met after John Franklin had married the widow

Hubbard in 1753. Caty and Jane were some 20 years apart in their age, and relationship was, in a way, that of parent and child, Caty calling her "my mama and friend."

The Lipit-Ishtar Law Code

Francis R. Steele

University Museum

University of Pennsylvania

Only last March four tablet fragments in the Nippur Collection of the University Museum were identified as parts of a single tablet upon which had been inscribed a law code in the Sumerian language. This code proved to be nearly two centuries older than the celebrated Babylonian law code of Hammurabi. Heretofore, it has been generally held that the Hammurabi law code was the oldest in the world, and credit for the codification of law has been assigned to this famous king of the First Dynasty of Babylon. Now, however, we find that Hammurabi had a predecessor in a relatively little-known king of a slightly earlier period, Lipit-Ishtar, king of the city of Isin.

It is the purpose of this paper to describe the discovery, identification, and reconstruction of the Sumerian law code from the four tablet fragments with the assistance of other tablets containing excerpts of Sumerian law from the same code. A brief statement of the historic situation, based upon the revised Babylonian chronology, follows in order to place the two law codes in their proper places in time. Then the interrelationship of the Babylonian and Sumerian law codes is discussed and illustrated by passages from the respective codes. Finally, the significance of the new law code for the social history of southern Mesopotamia is pointed out.

Why Union Education?—Aims, History and Philosophy of the Educational Work of the ILGWU

Mark Starr

International Ladies' Garment Workers' Union

The International Ladies' Garment Workers' Union was chartered by the national American Federation of Labor in 1900 to organize the makers of women's garments. It now has over 400,000 members in 435 locals and is strongly established in most of the metropolitan areas of the United States and Canada. The union's educational work and its motivating philosophy show three distinct phases:

(1) Agitation for remedial action against sweatshops to give the garment workers confidence in their own union solidarity and united action. In this the ILGWU received the support of progressive politicians and social, civic, and religious groups. Since the union was largely composed of immigrant workers, the early formal classes were largely in English, citizenship, and history, orienting them to the American scene.

(2) When the union's struggle for recognition as a nationwide force in the industry had been won, there was a shift in its educational activity to establish a relation of understanding with the community and a participation in varied forms of welfare work. In the New Deal period a rapid increase in membership of American-born youth with high school background and the union's enlarged financial resources made it

both necessary and possible to strengthen the relationship of the union to other sections of the community. This community activity was especially strengthened in wartime. The ILGWU continued its attempt to secure cooperation of outstanding forward-looking men and women in improving industrial conditions.

(3) Current union education has a threefold task:

(a) Union self-education for new members and the compulsory educational qualifications for its officers; and departments in research and industrial engineering to estimate trends and techniques in the garment industry to guide the policy of the union.

(b) The provision of social, recreational, and cultural opportunities directly, where necessary, but through community agencies wherever possible.

(c) The promotion of the study of industrial relations in conjunction with institutions of higher learning as a reflex of efforts in labor-management cooperation and the provision of health and welfare benefits.

The ILGWU aims by its educational work to replace social illiteracy by civic and political maturity and by the preparation and training for living in its broadest social implications.

Cepheids of the Small Magellanic Clouds

Harlow Shapley

Harvard College Observatory

and

Virginia McKibben Nail

Peculiar differences, in basic physical operations, from one galaxy to another are again suggested by current studies of Cepheid variables in the Large and Small Clouds of Magellan. Forty-nine new photographic light curves are presented, with observations based on a homogeneous magnitude system.

Anomalous characteristics appear among the hundreds of variables in these nearest of external galaxies. The Large Cloud appears to follow our own Galaxy's pattern in the distribution of period lengths. There are periods of all values from about 2 days to 100 days, with a maximum frequency near 4 days. In the Small Cloud, which is distant from the Large Cloud only about 10 kiloparsecs, and is similarly irregular in form, the maximum frequency is around 2 days—in fact, most of the periods of Cepheids in the Small Cloud fall in the interval of period lengths that is almost completely avoided by Large Cloud Cepheids.

Some additional results are now reported that again emphasize deviations from the conveniently assumed large-scale uniformity of the laws and operations of nature throughout the Metagalaxy. In the Magellanic Clouds, Cepheids of about 8-day period lengths have single maxima; in the Milky Way many of that period length have double maxima. But in the Clouds those with periods from 9 to 11 days mostly have double maxima, although in that interval the Milky Way Cepheids have single maxima. That is, there is a shift of 2 days in the appearance of this significant phenomenon.

The importance of these and similar results lies in their indication of cosmic nonuniformity, either in the distribution of the original chemical elements out of which stars are formed or in the hydrodynamical operations of a pulsing star, depending on position in space or environment.

The Freedmen and the Slaves of God

William Linn Westermann
Columbia University

In First Corinthians 7 the Apostle Paul set forth the equalitarian principle which he thought should prevail in the new Christian groups. A slave who was baptized became a "freedman of the Lord." A free man who accepted Christianity became a slave of Christ. This meant that differences in status could have no significance within the Christian communities. Each convert had been bought at the price of Christ's sacrifice; and each one was to submit himself to God as a slave, whatever the actual social status which he had at the time of his conversion. At times Paul also speaks of himself, with genuine humility, as a slave of God; and this idea has come down in the

Church, in the bulls of the Popes, in which they speak of themselves under the humble title of *servus servorum Dei*.

The Greek temple organizations might have slaves; but the Greek gods did not have them. The Semitic gods, on the other hand, *did* have slaves. In the Greek city-states, particularly at Delphi, a system had arisen by which a slave might redeem himself through a trust purchase made by the god. When the god bought him, automatically the slave became free because the god had no slaves. The new freedman often contracted to serve his former owner for a time. This form of service, and the labor contract which covered it, was called a *paramone*. Paul had this legal formula of the Greek world in mind when he spoke of the freed man of the Lord. The idea, however, of the free man who became the slave of God was not Greek but came from the Semitic background of Saul of Tarsus.

Obituary

Ellsworth Huntington

1876-1947

Yale's Prof. Huntington was such an earnest, energetic searcher after truth, so desirous of making the best possible use of available information, so bold in presenting challenging hypotheses, and so skillful as a writer that his passing will be widely mourned. His early geological work was so notable that he was starred as a distinguished geologist in *American men of science*. He received medals from leading British, French, and American geographical societies for geographic exploration in little-known areas in Asia, and the "Distinguished Service to Geography Award" of the National Council of Geography Teachers. He served as president of the Association of American Geographers, the Ecological Society of America, and the American Eugenics Society, and as associate editor of the *Geographical Review*, *Economic Geography*, *Ecology*, and *Social Philosophy*. Numerous historians, economists, anthropologists, meteorologists, climatologists, physicians, and many others have frequently cited one or more of his numerous publications and acknowledged his stimulating influence in their fields. His several geographic textbooks have been widely used, serving students on all levels from grammar school to postgraduate work. An extensive investigation made a decade ago revealed that he was the most widely known American geographer—known not only by people interested in the earth sciences but by educated people generally. Several of his books have been translated into other languages.

Huntington is known especially for his researches in seven great fields and for the presentation of findings

so attractively that reading is a pleasure. His prolonged studies of past climates established that there have been highly significant changes of climate and that these were pulsations, not cycles or progressive. He proved by numerous factual studies that climatic conditions have profound influence upon man and his culture, upon other forms of life, and upon geologic processes. He also proved that the changes of weather, although brief and erratic, affect man's health, attitudes, and achievements and even his energy and longevity. He assembled convincing evidence that the distribution of "civilization" over the earth corresponds with that of climate. The climate best suited for intellectual activities is one having frequent changes of weather, well-marked seasons, and enough warmth and rainfall to permit extensive agricultural production, but not too much warmth. He concluded that temperatures above 68° are uncondusive to thought, while frequent drops below 50° are distinctly stimulating. His theories that changes in the sun are major causes of terrestrial climatic changes and that frequent, relatively intense, cyclonic disturbances, "storms," are conducive to intellectual progress have stimulated much discussion and ever-widening adoption. One of his several stimulating theories is that not only has civilization shifted northward into cooler climates as mankind has advanced in his culture, but for each of the chief crops and types of farm animals there has been a northward improvement (in the Northern Hemisphere) in yield and quality almost to the poleward margin of the crop's or animal's range.

Huntington, although sometimes considered to be an "extreme environmentalist" by persons who have not read his works carefully, was keenly aware that the

physical environment generally is of secondary or indirect significance for mankind. The great influences of selective migration and of the intermarriage of certain types of people are stressed in his *The character of races* (1924) and elaborated upon at length in *Mainsprings of civilization* (1945). In his *Human habitat* (1927) this subject is attractively and briefly presented.

Huntington was the author or co-author of more than 25 volumes, contributed a chapter to each of about 20 other books, and wrote more than 130 articles, more than a score of which were in magazines of wide circulation. His books include *Explorations in Turkestan* (1905), *Pulse of Asia* (1907), *Palestine and its transformation* (1911), *The climatic factor as illustrated in arid America* (1915), *Civilization and climate* (1915, 1924), *World power and evolution* (1919), *The red man's continent* (1919), *Principles of human geography* (1920-1940), *Principles of economic geography* (1940), *Climatic changes: their nature and causes* (1922), *Earth and sun* (1923), *The character of races* (1924), *West of the Pacific* (1925), *Quaternary climates* (1925), *Pulse of progress* (1926), *Builders of America* (1927), *Weather and health* (1930), *Living geography* (1932), *Tomorrow's children* (1935), *After three centuries* (1935), *Season of birth* (1938), and *Geography of human affairs* (1947).

Mainsprings of civilization (1945) is a monument not only to his exceptional erudition, energy, and persistence but to American science. Indeed, *Time*, in its recent obituary, characterizes it as surpassed only by Toynbee's classic in breadth of scholarship, wide interest, and literary attractiveness.

Dr. Huntington was born in a manse in Galesburg, Illinois, went to southwestern Asia upon graduating from Beloit College in 1897, returned again to Asia for further extended exploration after two years at Harvard (1901-03), and carried on years of field research in southwestern United States and Mexico. Later he did extensive field work in Europe, Africa, Australia, and South America. No other geographer has made such prolonged and varied foreign field studies, and none has shared his observations so attractively. *West of the Pacific* is considered by some competent persons as a "gem" of descriptive geography, but most of his works include effective descriptions. His death, from a heart attack on October 17, 1947, terminated his work on *The pace of history*, a supplement to *Mainsprings of civilization*. Undoubtedly Huntington's influence will be considerable as long as our civilization continues.

STEPHEN S. VISHER

Indiana University, Bloomington

NEWS and Notes

Bernard F. Riess, professor of psychology, Hunter College, has been appointed research associate, Department of Animal Behavior, American Museum of Natural History. Dr. Riess, who is spending a year at the Museum as a Guggenheim Fellow, is investigating biochemical factors affecting behavior in mammals.

Ernst Antevs, formerly of Harvard University and the Carnegie Institution of Washington, has been appointed research associate in glacial geology, Chicago Natural History Museum, and **Ch'eng-chao Liu**, professor of zoology, West China Union University, Chengtu, has been appointed research associate in the Division of Reptiles.

Jay McLean has resigned as associate professor of surgery, Ohio State University, to become director, Bureau of Cancer Control, Health Department

Government of the District of Columbia, Washington, D. C.

Hayse H. Black, formerly officer in charge, U. S. Section, International Joint Commission Boundary Water Pollution Investigation, Detroit, has been appointed associate professor of sanitary engineering, Department of Civil Engineering, State University of Iowa.

Laurence H. Snyder, dean of the Graduate College, University of Oklahoma, has been giving talks at various institutions on the general subject of human and medical genetics. On October 23 he spoke before the student body at Hollins College, Virginia; on October 28, before the Fort Henry Academy of Medicine, Wheeling, West Virginia; on December 5, before the Terre Haute Academy of Medicine; and on December 11, at the annual initiation ceremonies of Phi Sigma at the University of Texas.

Willard Rouse Jillson, geologist and engineer, Frankfort, Kentucky, and formerly director of the Kentucky Geological Survey, has been appointed professor and head, Department of Geology, Transylvania College, Lexington, Kentucky.

Walter Buehler, a graduate of Purdue University and past-president of the American Wood Preservers Association, has been appointed consultant in wood technology and preservation, School of Forestry, University of Florida.

Sidney Q. Janus has been appointed part-time professor, and **A. S. Glickman** as instructor, in the Department of Psychology, Georgia School of Technology.

Grants and Awards

Six new grants for research in mental health under the National Mental Health Act have been recommended by the National Advisory Mental Health Council and approved by Thomas Parran, Surgeon General, U. S. Public Health Service. Institutions receiving the grants, and their project directors, follow: University of California, Berkeley, **Karl M. Bowman**, professor of psychiatry and medical superintendent, and **Jurgen Ruesch**, research psychiatrist and lecturer in psychiatry, Langley Porter Clinic; Columbia University, College of Physicians and Surgeons, **Abner Wolf**, associate professor of neuropathology; The Roscoe B. Jack-

son Memorial Laboratory, Bar Harbor, Maine, **J. P. Scott**, chairman, Division of Behavior Studies; Mu Iota Sigma Fraternity, Illinois School for the Deaf, Jacksonville, Illinois, **M. Arline Albright**, associate professor of education and psychometrist, and president, Mu Iota Sigma Fraternity, Milwaukee, Wisconsin; Wayne University School of Public Affairs and Social Work, Detroit, Michigan, **Fritz Redl**, professor of social work, and **Ronald Lippitt**, director of research, Research Center for Group Dynamics (Massachusetts Institute of Technology); and Wesleyan University, Middletown, Connecticut, **David C. McClelland**, assistant professor, Department of Psychology.

Charles Clemon Deam, Indiana, botanist, received the second Mary Sope Pope Medal of the Cranbrook Institute of Science, December 18, in recognition of a lifetime of collecting and publishing on the Indiana flora, and especially for his work, *Flora of Indiana*.

Colleges and Universities

The Medical School of the University of Birmingham, England, will conduct a four-week Post-Graduate Summer School, July 10-August 7, 1948, which is intended for American, British, and European students, both men and women, who have done advanced study in the appropriate fields of science and medicine. The session, to be divided into two terms of two weeks each, will offer the following main courses: (A) The Physiology of the Sex Hormones, four weeks, 16 seminars; (B) Normal and Abnormal Fat Absorption in Experimental Animals and Man, first term, 8 seminars; and (C) Lipoprotein Association in Biological Systems, second term, 8 seminars. Several short courses will be arranged in each term, as well as a series of visits to leading hospitals, medical centers, and places of historical interest in the vicinity. Opening speakers at seminars will include S. Zuckerman, professor of anatomy, University of Birmingham, and A. C. Frazer, professor of pharmacology, University of Birmingham. J. J. Elkes, lecturer in pharmacology, and P. L. Krohn, lecturer in anatomy, both of the University, will be deans of the School for the first and second terms, respectively, while Sir Leonard Parsons, dean, Faculty of Medicine, and emeritus professor of

pediatrics, will be president of the School. Cost of the four weeks will be \$96 for tuition, and \$80 for board and room in one of the University residence halls. American applications should be submitted by March 15 to the Director, Institute of International Education, 2 West 45th Street, New York 19, New York, marked "Birmingham University Medical Summer School."

Meetings

The 1948 annual meeting of the American Institute of Chemists will be held Saturday, May 8, 1948, at the Hotel Pennsylvania, New York. The program, to be announced later, will commemorate the Silver Anniversary of the Institute, which was founded in New York in 1923.

The First International Poliomyelitis Conference will be held at the Waldorf-Astoria Hotel, New York, July 12-17, 1948, under the sponsorship of the National Foundation for Infantile Paralysis. More than 60 foreign countries are expected to be represented by officials who will be asked to present summarizations of the problems of poliomyelitis in their countries at a special session which will be in charge of Thomas Parran, Surgeon General, U. S. Public Health Service. Basil O'Connor, president of the Foundation, will be the official host to the delegates, while Hart E. van Riper, medical director of the Foundation, has been appointed general chairman. Scientific and technical papers on research and treatment of poliomyelitis will be presented by professional authorities from this country and abroad, and in addition, the program will include panel discussions on the various subjects. The program is being arranged by an advisory board including Irvin Abell, clinical professor of surgery, University of Louisville; Morris Fishbein, editor, *Journal of the American Medical Association*; David Lloyd, associate member, Rockefeller Institute for Medical Research; Kenneth Maxcy, professor of epidemiology, Johns Hopkins University; Rustin McIntosh, professor of pediatrics, Columbia University; Frank Ober, professor emeritus of orthopedic surgery, Harvard University; and Thomas Rivers, director, Hospital of the Rockefeller Institute for Medical Research.

In addition to the sessions, there will be a scientific exhibit section, demon-

strations of muscle testing and treatment procedures, and a film program. Coordinating this phase of the Conference will be an advisory committee consisting of Thomas G. Hull, director of scientific exhibits, American Medical Association, and Charles F. Branch, director of scientific exhibits, American College of Surgeons.

Conference headquarters have been established in the Waldorf-Astoria under the direction of Stanley E. Henwood, of Chicago, who has been appointed executive secretary.

An International Rheological Congress will be organized, on the initiative of the British Rheologist's Club, by a group of Dutch rheologists under the sponsorship of the Joint Committee on Rheology of the International Council of Scientific Unions and the Rheological Committee of the Royal Netherlands Academy of Sciences at Amsterdam. The Congress will probably meet in Scheveningen, Holland, September 21-24, 1948. The provisional program follows: Tuesday morning, general lecture on Recent Developments in the Theory of Viscosity; afternoon, communications from the Joint Committee, and general lectures on Nomenclature, and Fundamental Experimental Methods; Wednesday morning, sectional meetings; afternoon, lectures on Solutions of Macromolecular Substances, and Russian Work on Rheology; Thursday morning, sectional meetings, and lecture on Relations Between Stress and Strain in Complicated Systems; afternoon, lectures on Abnormal Substances and Abnormal Phenomena of Flow, and Psychophysical Aspects of Rheology; evening, informal dinner and entertainment; Friday morning, sectional meetings, and lecture on Rheological Problems in Biology; afternoon, lecture on Rheology in Industry, and communications from the Joint Committee; Saturday, and possibly the following Monday, excursions if sufficient interest is shown.

Those wishing to present a paper should communicate with the first secretary, Dr. R. Houwink, Rubber-Stichting, Julianalaan 134, Delft, Holland. Manuscripts must not exceed a maximum space of 3,000 words, formulas and diagrams included, and must be received before May 1, 1948. The maximum time to be allowed each paper in the sectional meetings will be about 30 minutes, and it is expected that the speaker will give

only a short account of his paper so that ample time will be available for discussion. Reprints of all papers will be forwarded to participants at least a month in advance of the Congress. Registration fee will be about \$5.00.

The organizing Committee, headed by J. M. Burgers, chairman, and Dr. Houwink (Rheological Committee of the Netherlands Academy of Sciences), consists of H. Kramers (Netherlands Physical Society), A. J. Staverman (Netherlands Chemical Society), R. N. J. Saal (Section of Oil Technics, Royal Institution of Engineers), A. van Rossem (Society for Materials), and H. C. den Daas, assistant secretary.

The Indiana Academy of Science held its 63rd annual meeting, October 16-18, at Ball State Teachers College, Muncie, Indiana. The annual banquet was held Friday evening, followed by an address by J. F. Mackell, president, on "Graduate Degrees for College Science Teachers." An anonymous donor has made possible the establishment of an annual award to be given in each of two sections of the Academy for outstanding work during the previous 5 years. The awards will bear the names of past-presidents. At this meeting Ray C. Friesner, head, Department of Botany, Butler University, was awarded the John M. and Stanley Coulter Prize in Botany, and J. Paul Scott, formerly of Wabash College, was granted the David Starr Jordan Prize in the Zoology Section. Approximately 100 papers were read at the 12 divisional meetings on Friday and Saturday.

At the close of the business meeting Winona Welch, DePauw University, was elected president; Mason Hufford, Indiana University, vice-president; O. B. Christy, Ball State Teachers College, secretary; Donald E. Miller, Ball State Teachers College, assistant secretary; W. P. Morgan, Indiana Central College, treasurer; P. D. Edwards, Ball State Teachers College, editor; and William A. Daily, Butler University, press secretary.

The press secretary has announced that the 1948 annual fall meeting will be held October 28-30 at Indiana University, Bloomington.

T. H. Manninen, as the newly appointed manager of development, U. S. Industrial Chemicals, Inc., will supervise all pilot plant and engineering develop-

ment activities of the company. Dr. Manninen has been with USI since 1937.

Centro Research Laboratories, Briarcliff Manor, New York, have recently become associated with Chemisch-Technisch Adviesbureau, Haarlem, Holland, which is directed jointly by J. Rinse, a leading research worker on the continent in the field of organic coatings, plastics, pigments, and associated materials, and W. Dorst, a specialist in plant layout, especially design and the practical application of paints and other coating materials. The Holland plant has carried out extensive studies in the chemistry of both raw materials and intermediates such as the polymerization, dehydration, vinylation, and blowing of linseed, soybean, tung, oiticica, and castor oils, resin modification, chlorination of rubber, the formulation of fungicides and preservatives, and the products of pigments and other raw materials. The development of new and interesting products such as metal putties, plastic coatings for concrete, and grease-resisting and shrinkage-controlled textiles is anticipated by Centro as a result of this new association with the Dutch concern.

Recent Deaths

G. H. Hardy, 80, formerly Sadleirian professor of mathematics, University of Cambridge, England, died on December 1 in Cambridge. Prof. Hardy was recently named to receive the Copley Medal of the Royal Society.

S. Lawrence Bigelow, 77, professor emeritus of general and physical chemistry, University of Michigan, died December 3 in West Hartford, Connecticut.

Wesley G. France, 55, professor of colloid chemistry and electrochemistry, Ohio State University, died December 4 of a heart attack.

Rollins A. Emerson, 74, head of the Department of Plant Breeding at Cornell University from 1914-1942, and dean of the Graduate School from 1925 to 1930, died December 8 at Ithaca after a long illness.

Lord Rayleigh, 72, onetime president of the British Association for the Advancement of Science, emeritus professor of physics and chairman of the governing body, Imperial College of Science, died

December 13 at his home near Witham, Essex, following a heart attack.

Earl S. Johnston, 58, chief of the Division of Radiation and Organisms, Smithsonian Institution, died December 17 at his home in College Heights, Maryland, after an illness of several months.

The American Museum of Natural History is sending the first major American expedition to the Cape York Peninsula, Australia, in March, for a 6-month zoological and botanical exploration. The 1948 Archbold Cape York Expedition will be led by **Leonard Brass**, Museum botanist, who has already left for Australia, and include as party members, **G. H. H. Tate**, curator, Department of Mammals, **Hobart M. Van Deusen**, also of the Department of Mammals, and **G. M. Tate**, who will be in charge of transportation, and collection of insects, reptiles, and amphibians. This is one of a series of biological investigations in Australian and New Guinea areas conducted by Archbold Expeditions, an organization established by Richard Archbold and affiliated with the Museum. This expedition will concentrate on the collection of mammals and plants, although amphibians, reptiles, and insects will also be sought for the Museum's collections. The group expects to discover a number of new species of both mammals and plants. Seeds of palms and other ornamental plants will also be collected for cultivation at the Fairchild Tropical Garden in Florida. It is believed that the most interesting and profitable areas for collecting will lie in the northern part of the Peninsula, between Coen and Somerset. The group will establish a series of bases connected by water transport on the eastern coast of the Peninsula, and will cover not only the tropical rain forest areas there, but work into the western open Eucalyptus forest territory, making it possible to study a wide range of biological environments of the Peninsula. The plant collections will go to the Arnold Arboretum, which is giving financial support to the project.

Electronic engineer and physicist positions are available in several Federal research laboratories, including the U. S. Navy Underwater Sound Laboratory, Fort Trumbull, New London, Connecticut; Naval Research Laboratory

Field Station, 470 Atlantic Avenue, Boston; and the Cambridge Field Station, Watson Laboratories, Air Material Command, 230 Albany Street, Cambridge, Massachusetts. Salaries range from \$3,397 to \$8,179 per year. To qualify, applicants must have completed (a) a full four-year college course with major work in physics, mathematics, or engineering science, or (b) at least four years of progressive technical experience in engineering or physics, or (c) any time-equivalent combination of education and experience. All applicants for the position of physicist must have completed 24 college semester hours in physics. In addition, all applicants must have had from one to four years of progressive professional experience in the appropriate field. Graduate study may be substituted for experience up to a maximum of two years of experience. No written test is required. Full information and application forms may be secured from most first- and second-class post offices, from civil service regional offices, or from the U. S. Civil Service Commission, Washington 25, D. C. Interested persons should ask for Announcement No. 1-34(47). Applications will be accepted until further notice by the executive secretaries of the Boards of U. S. Civil Service Examiners at the laboratories named above.

TECHNICAL PAPERS

The Common Cold: A Note Regarding Isolation of an Agent¹

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For the past few months studies of the common cold have been in progress by the National Institute of Health. The infectiousness of nasal washings from cases of the disease and from laboratory materials has been investigated by intranasal inoculations of adult male volunteers² in a nearby correctional institution. Attempts to isolate and cultivate an agent or agents have been carried out at the Institute.

On January 13, 1947, nasal washings in sterile skimmed milk (1, 4, 10) were obtained from an individual within 24 hours of onset of cold symptoms. After laboratory studies to rule out, in so far as practical, the presence of dangerous pathogens, the unfiltered washings were given intranasally to 5 volunteers who had been in strict isolation in the institution hospital for 6 days. All subsequent groups had preliminary isolation periods of 6-8 days. After an incubation period of from 36 to 48 hours, all 5 volunteers developed symptoms and signs of minor upper-respiratory infection with considerable variation in severity.

All 5 complained of fatigue, nasal obstruction, and frequent expectoration. Three complained also of sneezing, headaches, coughing, and burning, watery eyes. Two experienced prodromal chilly sensations and hot flashes. Objectively, hyperemic obstructed nasal passages, red throats with promin-

ent lymphoid follicles, and large quantities of postnasal mucus for 2-5 days were evident. Slight temperature elevations between 99° and 99.6° developed after onset. One volunteer suffered all the above complaints to a more severe extent, and in addition developed moderate rhinitis, a mild laryngitis for a day, moderate pharyngitis, episodes of sharp, sticking pains substernally for 3 days (X-rays normal), and a temperature elevation to 102° F. for a day.

Nasal washings in sterile skimmed milk taken within 24 hours of onset of symptoms in this volunteer were inoculated into embryonated hens' eggs along with 1,000 units of penicillin and 100 units of streptomycin.

After two passages by way of the allantoic cavity, there was an unusual number of deaths of embryos between the 4th and 6th days following inoculation. The fluids from these proved bacteriologically sterile, and several substrains were established. One of these substrains was tested in the 4th passage in human volunteers and produced a mild illness reminiscent of the original volunteer group. In fluids from the 5th egg passage, however, no obvious clinical disease was produced.

A second substrain was passed blindly every 3 days. After 8 passages allantoic fluids were tested in a group of volunteers with a failure to produce disease.

A third substrain was carried through 4 allantoic passages, transfers being made from the 5th to the 7th day following inoculation. Occasional embryos died between the 3rd and the 5th days. After the 4th passage a pool of allantoic fluid, when tested in volunteers, produced mild coryza and a slight elevation of body temperature. An equal number of volunteers given normal allantoic fluid remained well.³ Two substrains were started from the allantoic fluid pool which produced the

¹ In this brief paper no attempt is made to review the literature. Some previous reports concerning the isolation of a possible agent or agents are listed under References (2, 3, 5-9).

² Certain criteria have been established for the selection of volunteer participation in the study. Volunteers must be healthy, physically fit, adult males between the ages of 20 and 40 years, who have given no history or physical findings of chronic sinusitis, allergic rhinitis, tuberculosis, or recent pneumonia. They must have no history of any respiratory infection for the past 6 weeks and no nasopharyngeal abnormalities.

We wish to acknowledge the generous cooperation of the volunteers as well as that of the Department of Corrections, District of Columbia, in making its facilities available for the human studies of the common cold.

³ Volunteers, hospital attendants, and the nurse are purposefully kept unaware of the nature of the inoculum and the identity of the controls. The volunteers frequently are apprehensive for the first 24-48 hours following intranasal inoculation. In most control groups some mild and transient nasal irritative phenomena are observed during this period. Between 25 and 50 per cent of the controls have occasional sneezing and slight nasal obstruction with some hyperemia and swelling of the nasal mucosa. A few have complained of a headache. In several controls a brief temperature rise has occurred.

mild illness. Both of these were tested in groups of 7 human volunteers. All 7 in each group developed signs and symptoms comparable to the upper-respiratory infections seen in the original donor and the first volunteer group. However, in this total of 14 volunteers the illness was more severe and of longer duration, persisting 7-11 days. After 7-24 hours incubation, all complained of dry, irritated throats (without objective pharyngitis) and exhibited malaise out of proportion to physical findings. Within a few hours nasal obstruction and postnasal discharge with frequent expectoration developed and remained prominent during the course. All complained of frequent supraorbital headaches, moderate sneezing, and an infrequent, mild, nonproductive cough. Hoarseness was evident in 9, one of whom became aphonic for about 12 hours. Nine complained of burning, watering eyes (with mild conjunctivitis objectively) and vague chest aches without significant X-ray findings. Intervals of profuse serous rhinitis were observed in 5. Early in the illness, 5 complained of chilly sensations and hot flashes. Objective signs of pharyngitis, lymphoid follicular prominence, hyperemic obstructed nasal passages, and profuse postnasal discharge were observed in all. Temperatures between 99° and 100° F. occurred at irregular intervals in 12 volunteers. No significant urine or white blood count changes were apparent. An equal number of controls selected at random from among the volunteers received normal allantoic fluids with and without chorioallantoic membranes at the same time and under conditions identical with those receiving infectious material. These men remained well and showed no change in diurnal temperature variations.

Sixth-passage allantoic fluid produced a moderately severe upper-respiratory infection of 8-11 days duration in 15 of 16 volunteers inoculated intranasally.

Seventh-passage allantoic fluid produced a similar clinical disease in 14 of 16 volunteers. No decrease in severity of infection has been evident.

To date, 8 groups of volunteers totaling 60 individuals have been inoculated with allantoic fluids or fluids plus membranes from embryonated hens' eggs inoculated with the agent. Of the 60 individuals, 57 have developed a characteristic syndrome that has been consistent through the 8 groups. Simultaneously, 8 groups of controls totaling 48 individuals have been inoculated with noninfected allantoic fluids or fluids plus membranes. Except for mild, transient, nasal irritative phenomena in from 25 to 50 per cent of these, they have remained well.

The exact nature of the agent, whose presence in the allantoic fluids and in macerated allantoic membranes plus fluids was demonstrated by producing signs and symptoms in the human volunteers, has not been defined. Bacteriological cultures of allantoic fluids and of fluids plus macerated membranes have been sterile in thioglycollate and Casman's blood agar media. Preparations stained by the Giemsa and Macchiavello techniques, when studied microscopically have failed to reveal bodies suggestive of bacteria or the larger viruses.

The material has so far failed to produce symptoms in several strains of mice, hamsters, rats, cotton rats, guinea pigs, and rabbits. Allantoic fluids have failed to agglutinate chicken red cells, and the volunteers have shown no rise in antibody to either influenza A or B.

The infectiousness of the material is preserved, at least for several weeks, when rapidly frozen at -70° C. and stored in the frozen state at -50° C. Preliminary electron microscopic

observations carried out by R. W. G. Wyckoff have shown characteristic particles in some active preparations which have not thus far been seen in preparations from normal allantoic fluids or from fluids of eggs inoculated with normal fluids. These particles are of the same general size as viruses of the influenza type but are readily distinguishable from them.

The substrains have been carried through several additional serial passages. Deaths of the embryo are quite infrequent, and grossly little, if any, change is noted in the embryo or membranes. Further testing of the material in human volunteers will be done as well as more extensive laboratory investigations.

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The Effects of Vitamins on Phosphorus Metabolism in the Chick Embryo:

1. Vitamin D and the Utilization of Inorganic Phosphorus

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As a preliminary experiment in the biophysical use of radioactive and stable isotopes, we have undertaken studies to obtain quantitative data relative to the effects of vitamins on the metabolism of phosphorus compounds in the developing chick embryo. Our initial work has been with vitamin D.

The biochemical action of vitamin D has been extensively studied (1). The most attractive hypotheses as to its mode of action seem to be that the vitamin accelerates the conversion of organic phosphorus into inorganic (1, 4) or, at least, intensifies the turnover of phosphorus in the bone (5). This work was performed with immature animals. Our experiments indicate that the vitamin accelerates the turnover of phosphorus in the developing embryo.

Large brown eggs (New Hampshire) were injected with 0.1 ml. of an isotonic solution of NaH_2PO_4 with an activity of 639.5 counts/second, or 0.12 microcuries. (The efficiency of our Geiger counter equipment, as determined by daily checks with the Bureau of Standards sample of Radium D and E, No. 26, was 9.84 per cent.) A portion of the eggs also received 0.1 ml. of propylene glycol containing 20 units of vitamin D;

another portion, 0.1 ml. of propylene glycol only; and the final group, only the NaH_2PO_4 . The eggs were sealed with sterile paraffin, marked, and placed in a modified Buckeye incubator with temperature and humidity control. Thirteen-day embryos proved convenient for analysis with a relatively high (40 per cent) survival rate.

The viable embryos were lifted with sterile forceps, cleaned of adhering membranes, and dropped immediately into liquid air. They were ground to a powder in a chilled mortar and the powder extracted several times with cold 10 per cent trichloroacetic acid followed by extracts with cold 5 per cent acid. The scheme of separation followed, with some modifications, that of Hevesy, *et al.* (2). The acid extract was filtered into cold, concentrated sodium hydroxide. The resulting solution was divided into three parts. From one aliquot we determined the average acid-soluble phosphorus compounds;

TABLE 1

Fractions	Activity: $\left(\frac{\text{Average counts}/\mu\text{g of phosphorus}}{\text{Counts from 0.1 ml. of original NaH}_2\text{PO}_4 \text{ solution}} \times 10^{+5} \right)$		
	Group receiving P* and vitamin D	Group receiving P* and propylene glycol	Group receiving P* only
Average acid soluble.....	0.530	0.522	0.633
Inorganic P.....	6.03	19.70	5.69
Adenosine P + inorganic P.....	8.12	4.00	2.18
Creatine P + inorganic P.....	4.15	3.64	3.32
Phosphatide P.....	2.33	1.65	0.825
Residual P (nucleoprotein).....	3.02	2.10	0.285

P* = P^{32} in NaH_2PO_4 .

the second aliquot was precipitated with 25 per cent barium acetate at pH 6.5, the precipitate was washed with dilute barium acetate, centrifuged, and a part dissolved in cold nitric acid. The solution was treated with ammonium molybdate reagent. This precipitate consisted of the inorganic phosphorus. The remainder of the barium acetate precipitate was ashed and the phosphorus determined. This fraction consisted of inorganic phosphorus plus adenosine phosphorus. The third aliquot was hydrolyzed with normal hydrochloric acid and 0.1 normal ammonium molybdate for 30 minutes at 40° C. The phosphorus released from the organic compounds precipitated. This fraction consisted of inorganic phosphorus plus phosphocreatine. The residue from the acid extractions was treated with an alcohol-ether mixture. The filtrate contained the phosphatide phosphorus, and the residue gave the so-called residual phosphorus containing mainly nucleoprotein phosphorus.

The phosphorus was determined in a Coleman junior spectrophotometer at 650 $\text{m}\mu$, according to the procedure of Kitson and Mellon (3).

A summary of our results is given in Table 1. Detailed analyses for each embryo and for anatomical parts of 18-day embryos will be presented at a later date.

Our data reveal that the group of 6 embryos receiving the P* and vitamin D shows a higher specific activity for all fractions except inorganic P and the average acid-soluble P than the group of 5 embryos which received P* and propylene glycol and the 2 embryos which received only P*. The inorganic

phosphorus value for the second group is inordinately high but we can discover no contamination or error. These average values support the contention that vitamin D accelerates the over-all metabolism of inorganic phosphorus in the developing chick embryo. The effect is most marked in the adenosine creatine, and phosphatide phosphorus fractions. Our data do not show any marked influence of vitamin D on the specific activity of the inorganic phosphorus fraction.

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Streptomycin Therapy in Experimental Tuberculosis of Guinea Pigs Infected Intracerebrally With Virulent Tubercle Bacilli¹

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Up to the present time published data is not available concerning the effect of streptomycin in animals infected intracerebrally with virulent tubercle bacilli. The following is a preliminary report of such an experiment.

Twenty tuberculin-negative female guinea pigs weighing between 800 and 1,000 grams each were inoculated intracerebrally with 0.05 cc. of a culture containing 0.0001 mg. (dry weight) of a 7-day growth of H37Rv organisms in Tween-albumin liquid medium of Dubos (1). A small drill hole was made through the posterodorsal part of the skull, just to one side of the midline, and the organisms introduced by means of a tuberculin syringe and a $\frac{1}{4}$ " No. 26 gauge hypodermic needle.

Ten animals were kept as untreated controls; the other 10 were started on streptomycin² treatment immediately after inoculation. They were given 4,000 μg at 7:30 A.M., 12:30 P.M., and 5:30 P.M., and 6,000 μg at 10:30 P.M., or a total of 18,000 μg /day. The drug was given either intramuscularly or subcutaneously.

During the third week after infection, both treated and control animals began to show signs of brain damage manifested by paralysis of the hindquarters, loss of equilibrium, and occasional convulsions upon stimulation. The control animals began to succumb on the 17th day, and all except one were dead by the 22nd day. The latter animal died on the 92nd day and developed increasing paralysis of the hindquarters for one week prior to death.

¹ This work was done under a grant-in-aid from the U. S. Public Health Service through the American Trudeau Society Streptomycin Program.

² Streptomycin was supplied through the courtesy of Dr. James Carlisle, of Merck and Company, Rahway, New Jersey.

On the 22nd day, at which time most of the treated animals were exhibiting the signs described above, the dose of streptomycin given to them was doubled, so that the animals received 36,000 μ g/day. Within a few days the treated animals began to show improvement. The convulsive attacks and paralysis disappeared rapidly, the loss of equilibrium more slowly. Except for two animals which died of intercurrent disease on the 5th and 10th days after infection, the treated guinea pigs all remained well thereafter. After the 58th day, streptomycin treatment was stopped in one-half the group (4 animals), the remainder continuing on treatment. Those animals deprived of streptomycin showed a gradual return of paralysis of the hindquarters, which became progressively worse until time of death. All 4 animals died from the 98th to the 132nd day after infection (an average of 58 days after treatment was stopped).

TABLE 1

Guinea pig No.	Days of treatment with streptomycin	Days of life
1	None*	21
2	"	17
3	"	92
4	"	19
5	"	19
6	"	19
7	"	18
8	"	19
9	"	22
10	"	21
1	5	5*
2	10	10*
3	58	132
4	58	127
5	58	109
6	58	98
7	173†	Living
8	173†	"
9	173†	"
10	173†	"

* Died of intercurrent disease.

† As of September 3, still living and being treated.

The 4 guinea pigs which were kept on treatment are still alive and well 173 days after infection.

All the treated animals responded to 5 per cent Old Tuberculin when tested intracutaneously 40 days after infection. The 4 remaining pigs are still skin positive at the time of writing this report, but the intensity of the reaction is declining. A summary of the time of death in relation to days of treatment is given in Table 1.

This experiment demonstrates that streptomycin administered intramuscularly or subcutaneously can effect inoculation tuberculosis of the brain in guinea pigs and cause improvement in the peripheral manifestations of such a lesion.

Previous experience in this laboratory has shown that an inoculum of attenuated or heat-killed tubercle bacilli equivalent to the infecting dose used in this experiment is not sufficient to produce skin hypersensitivity in guinea pigs. The results set forth in this paper, therefore, indicate that multiplication of the infecting organisms must have taken place in the guinea pigs despite the presence of streptomycin, and that it was only when the mechanisms of acquired immunity of the

animal came into play that the disease was held under control by the drug. Further data on this subject will be reported from this laboratory (2).

The intracerebral method of infection offers a rapid means of testing the *in vivo* effect of antituberculosis drugs.

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"Acid Phosphatase" Reactions in Peripheral Nerves¹

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The methods developed by Gomori (4) to reveal phosphatases in tissues have produced striking differentiations and are important additions to histologic technique. There is, for example, no simpler or more specific method for demonstrating the axis cylinders of nerve fibers than the acid phosphatase method following acetone fixation. The interpretation of the reaction, however, has presented difficulties so far as the nervous system is concerned. Heinzen (5) compared the acid phosphatase activity in the central and peripheral stumps of a series of transected nerves by standard biochemical methods and found increased activity in both stumps as compared with normal nerves. Histologic sections prepared by the Gomori method, on the other hand, showed a feeble reaction in the peripheral stump at a time when both stumps exhibited great activity *in vitro*. He suggested that the peripheral stump had suffered greater loss in staining capacity than the central between the time of removal of the tissue and the mounting of the sections.

Bodian (3) found the acid phosphatase activity in central stumps of transected nerves twice that of normal nerves and that of peripheral stumps 6 times as great. These biochemically determined differences could not be seen in sections prepared by the Gomori method. Bodian and Flexner then showed "that every step in the preparation of the histological sections for histochemical study sharply reduces the phosphatase activity as determined with the biochemical method."

We have studied the Gomori acid phosphatase reaction in peripheral nerves which had not been subjected to all the insults of histologic technique. Acetone-fixed nerves were teased in 80 per cent alcohol and transferred to the glycerophosphate-lead reagent buffered at pH 4.8 with molar acetate. After 2-18 hours at 38° C. they were washed thoroughly in distilled water and the lead visualized with ammonium sulphide.² Under these conditions the peripheral stump of a cat's sciatic nerve 16 days after transection appeared to the naked eye to be as intensely stained as the central stump. Under the microscope the normal fibers of the latter showed the usual precise staining of the axones; as in the normal controls, nuclei could rarely be demonstrated. Peripherally, where invasion of regenerating fibers had been prevented, all

¹ Aided by a grant from the Dr. Wallace C. and Clara A. Abbott Memorial Fund of The University of Chicago.

² Rinsing with 1 per cent aqueous acetic acid before the sulphide did not materially alter the picture.

types of nuclei reacted strongly, as did the degenerating remains of axones and myelin. The neuroma presented the same picture with many nuclei and, in addition, had the regenerating fibers differentiated in all detail. When strands of these same nerves were extracted with distilled water either *before* or *after* the glycerophosphate-lead treatment, the nuclei were the

irregularly over the larger myelinated fibers, thanks to the staining of small granules. Fig. 1 shows a Schwann cell after 1½ hours of incubation.

We have obtained this result in various normal nerves of cat, rabbit, rat, guinea pig, macaque monkey, and man.³ The promptness and regularity with which the Schwann cells

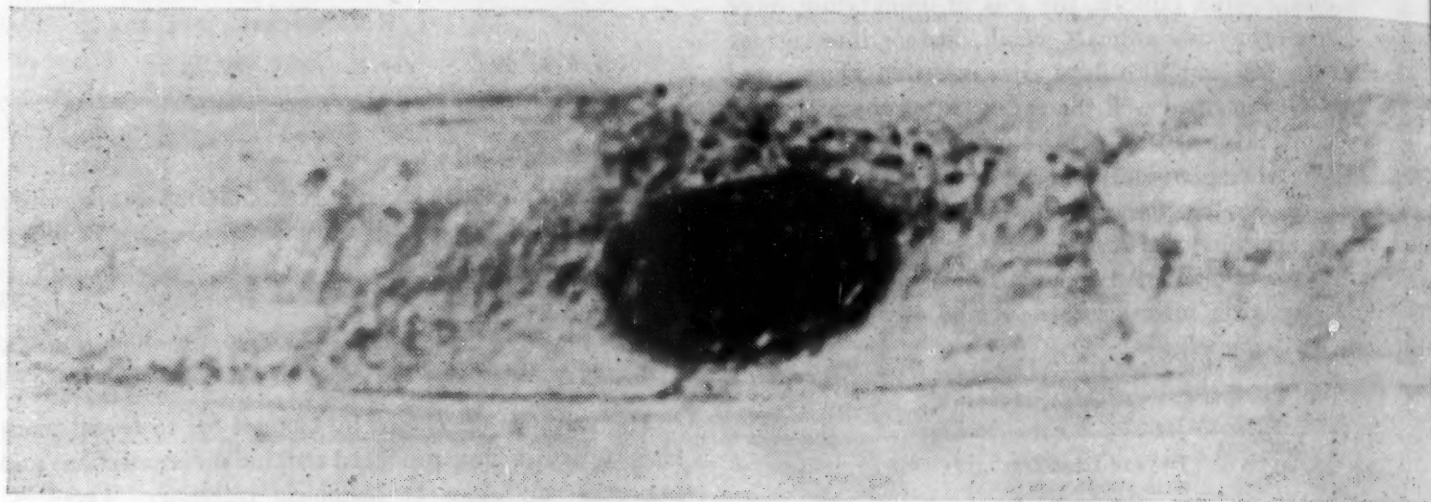


FIG. 1 Photomicrograph of cell of Schwann, viewed from above, showing stained granules in nucleus and cytoplasm. The cell appears irregularly branched. (From a bit of fresh sciatic nerve of a kitten placed directly into the glycerophosphate-lead reagent and incubated for 1½ hours. Photographed by R. D. Bensley from a teased nerve mounted in glycerine. (X 2,000.)

first to suffer. After extractions of 16–24 hours, which left the axones stainable, the nuclei no longer reacted at all. There is thus a differential susceptibility to washing with water which appears to be greater in paraffin material cut into thin sections, such as Heinen used, than it is in strands of teased fibers.

The water extractions provide an explanation of his discrepant results. They indicate that both enzymes and lead phosphate can readily be removed from the tissue. This would

the nuclei of other sheath cells react suggest that we are dealing with a highly specific reaction. There is no differentiation if glycerine is substituted for the glycerophosphate, if the fresh tissue is boiled for 5 minutes in distilled water,⁴ or if NaF is added to the reagent in a concentration of .005 M. If the reactions in fresh tissue are indeed indications of intracellular phosphatase activity, there is no reason for concluding that the various observed granules are actual sites of enzymatic

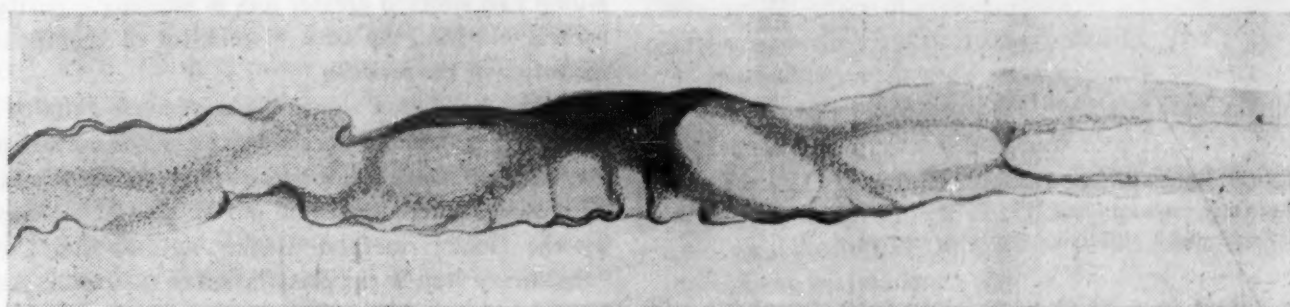


FIG. 2 Schwann cell in profile showing anastomosing cell processes which, when viewed on edge, appear as dark lines. (From the same nerve as Fig. 1 but incubated for 23 hours. Drawn by Agnes Nixon from a teased preparation in glycerine. (X 620.)

necessarily mean that both can move about freely within the tissue and that the Gomori method cannot be relied upon to furnish evidence as to the localization of acid phosphatase activity within the cell.

When acetone fixation was omitted and bits of fresh normal nerves were snipped off and placed directly into the glycerophosphate-lead reagent, the microscopic picture differed significantly in that the various types of nuclei were the dominant features. In the course of 25 minutes incubation at 38° C. an occasional nucleus appeared, and some cytoplasmic granules of the Schwann cells were sharply stained; a few nucleoli were darker than other structures. By 45 minutes practically all nuclei of sheath cells appeared filled with granules of uniform size, smaller than the nucleolus, which was darker. The cytoplasm of the Schwann cells could be followed as it spread

reactions. The reaction requires at least 25 minutes to attain microscopic visibility, and this affords ample time for the shifting of both enzymes and lead phosphate within the protoplasm. In this connection the findings of Owens and Bensley (7) on the drift and segregation of colloid particles in cells are particularly significant.

In the cat the cytoplasm of the Schwann cells develops a dark tinge in addition to the granules in the course of 18–24 hours of incubation. Cells such as those of Figs. 1 and 2 are present on every myelinated fiber midway between the nodes of Ranvier. The pictures resemble those obtained by Nemiloff

³ We are indebted for the human material to D. B. Phemister and Alex Brunswick.

⁴ This method of destroying phosphatase activity was suggested to us by W. L. Doyle.

in spinal root fibers of the cat stained intravitaly with methylene blue. In our material it is occasionally possible to follow the cell processes to the nodes, where there is usually an accumulation of cytoplasm studded with fine granules. Sometimes appearances like the spiny bracelets of Nageotte are sometimes differentiated. In time, the nuclei become opaque (Fig. 2) through the enlargement and fusion of the granules. Another phenomenon appears in the fresh tissue, and as promptly as the staining of the sheath cells. The ends of the axons react intensely where they were injured by pressure or electrical changes induced by metal instruments in the fresh tissue. Similar reactions to injury occur with methylene blue in living animals (2). The lead sulphide staining at sites of injury always occurs after boiling fresh tissues and has nothing to do with phosphatases. In addition, the normal myelinated fibers assume a uniform yellow tint, both in fresh and boiled material. This is presumably due to a diffuse adsorption of lead. That it is not due to staining with sulphides is indicated by the fact that a similar uniform staining occurs if the lead is visualized by means of a freshly prepared unoxidized aqueous solution of hematoxylin.⁵

If dry ice is applied to a living nerve *in situ*, all axones are revealed. When the frozen tissue is gradually thawed in chilled glycerophosphate-lead reagent and then incubated, all axones are as intensely stained as in acetone-fixed material. In addition, all nuclei react as in fresh tissue. In 1906 R. R. Bensley (1) showed that the supposed histochemical test for organically bound phosphorus was nothing more than a staining reaction. The Gomori phosphatase reactions likewise involve substitutions; the substance visualized is not the substance sought. Adsorption and diffusion may play significant roles. Until it has been shown that they do not, the specificity of the reactions must remain in doubt. It may be that both boiling and fluorides do more than merely inhibit enzymatic activity.

Certain findings indicate that the histologic acid phosphatase reactions are unreliable: (1) Both enzymes and lead phosphate can be leached out of, and hence moved about within, fixed tissues. This deprives the method of value for the localization of enzymatic activity. (2) All nuclei and the Schwann cell cytoplasm react promptly when fresh nerves are placed directly in the glycerophosphate-lead reagent. This does not occur after acetone fixation. (3) The axones of fresh myelinated fibers do not react unless they have been injured. Axones shriveled by freezing or acetone react strongly except at the nodes of Ranvier. (4) The reaction of injured tissue after the destruction of enzymatic activity shows that under certain conditions the lead of the reagent can be specifically adsorbed.

We are continuing our efforts to obtain differentiations in peripheral nerves with reagents which simulate the glycerophosphate-lead reagent but which contain no phosphate.

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⁵We owe this test to R. R. Bensley.

Polydactyly and Limb Duplication Occurring Naturally in the Tiger Salamander, *Ambystoma tigrinum*

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Few cases of polydactyly and limb duplication occurring in nature in *Ambystoma* have been reported in the literature (8). On the other hand, limb duplications following transplantation procedures in the laboratory are very well known from the work of Harrison and his students. Such anomalies may result from a variety of operative treatments, including the division of the limb rudiment (11), orthotopic and heterotopic limb transplantations (2, 3, 5, 7, 9, 12), and induction of supernumerary limbs by implantation of foreign tissue (1, 4, 6). The experimental production of limb duplications by methods other than transplantation techniques is still an unexplored field.

TABLE 1
POLYDACTYLY AND LIMB DUPLICATIONS IN *Ambystoma tigrinum**

Animal	Left hind limb	Right hind limb	Remarks
2	Normal	8 digits	Metamorphosed
3	6 digits	6 "	
4	Normal	6 "	
5	Main limb with 6 digits; extra limb with 12 digits	Main limb normal; small extra limb with 2 digits	
6	8 digits	8 digits	
7	Normal	7 digits plus 1 very small bud	Questionable mirror-image symmetry
8	Main limb with 7 digits; extra limb with 7 digits	8 digits	Questionable mirror-image symmetry
9	6 digits	6 "	
10	Normal	8 "	
11	6 digits	6 "	
12	9 "	9 "	
13	Normal	8 "	
14	Main limb normal; extra limb with 7 digits	7 "	
15	6 digits	8 "	
16	9 "	Main limb and extra limb fused with a total of 14 digits	
17	9 "	6 digits plus 1 small bud	Metamorphosed
18	6 " plus 2 buds	6 digits	"
20	Normal	6 "	
21	6 digits	Normal	

* Animals 2 through 18 collected as larvae, 1946 (117-144 mm. in length); 20 and 21 collected as adults, 1947.

The anomalies reported here in the tiger salamander, *Ambystoma tigrinum*, represent the first recorded case of mass polydactyly occurring naturally. The abnormal animals were collected in Muskee Lake (altitude, 8,300 feet) approximately 15 miles west of Boulder, Colorado. This and the surrounding lakes have been extensively studied in recent years, but the first polydactylous individuals were found in October 1946 (17 larvae) and in April and May 1947 (2 adults). The per-

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centage of abnormal individuals collected in the fall of 1946 was extremely high—of 19 animals collected, 17 showed polydactyly and limb duplication in some degree.

The abnormalities range from one extra digit to a complete extra appendage with supernumerary toes. The condition is found in both sexes, on either one or both sides of the animal,

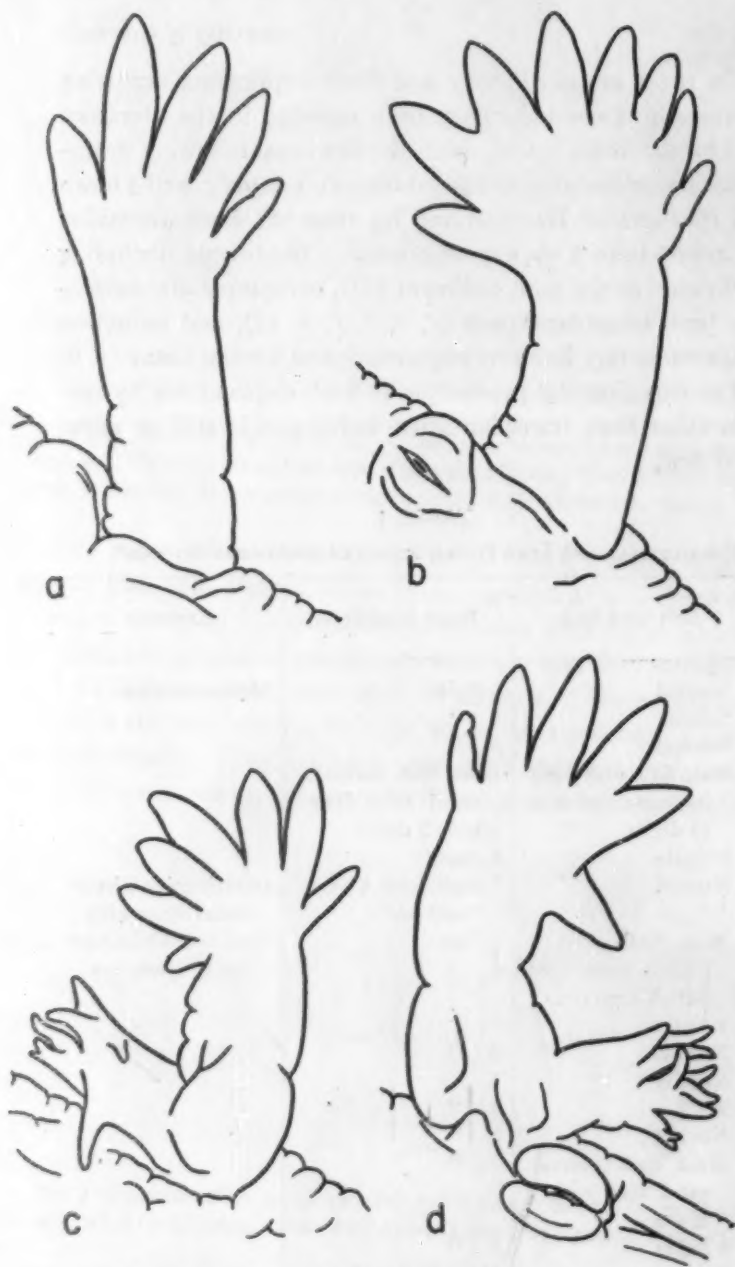


FIG. 1. Polydactylous hind limbs of *A. tigrinum* (2½X): (a) normal left limb, spec. #10, dorsal view; (b) 8-toed right limb, spec. #10, ventral view; (c) extra polydactylous left limb, spec. #8, lateral view; (d) left limb duplication, spec. #5, ventral view.

and is always limited to the hind limbs. A summary of the abnormalities occurring in larvae and adults collected both seasons is presented in Table 1. Outline drawings of polydactylous and duplicated limbs from three larvae are shown together with a normal limb for comparison (Fig. 1). The anomalous condition is not altered by metamorphosis of the animals from the larval to adult stage. Pigmentation of the extra digits is normal, and they appear in most cases to be fully functional. However, the supernumerary limbs seem to be more of an encumbrance than an advantage to the salamanders.

Polydactyly and limb duplications induced by transplantation methods usually show mirror-image symmetry between the supernumerary and the normal limb, although asymmetrical

exceptions have been noted, particularly by Swett (10). This symmetry holds for duplications in both the dorsoventral and the anteroposterior planes and is due, according to Harrison (6), to the development of more than one growth center, the main center reversing the symmetry of an adjacent center. It is of significance to point out that in the naturally occurring polydactylous series noted in this paper there are only two cases—and they are doubtful—of mirror-image symmetry (Fig. 2). The right hind limb of animal #7 has 7 well-formed toes plus a small bud, and we may be dealing with a duplication involving a reversal of symmetry. In animal #8 the supernumerary left limb can be interpreted as dorsoventral mirror-image symmetry, with, however, considerable modification due to different growth rates of the several digits. These two cases of questionable mirror-image symmetry are exceptions

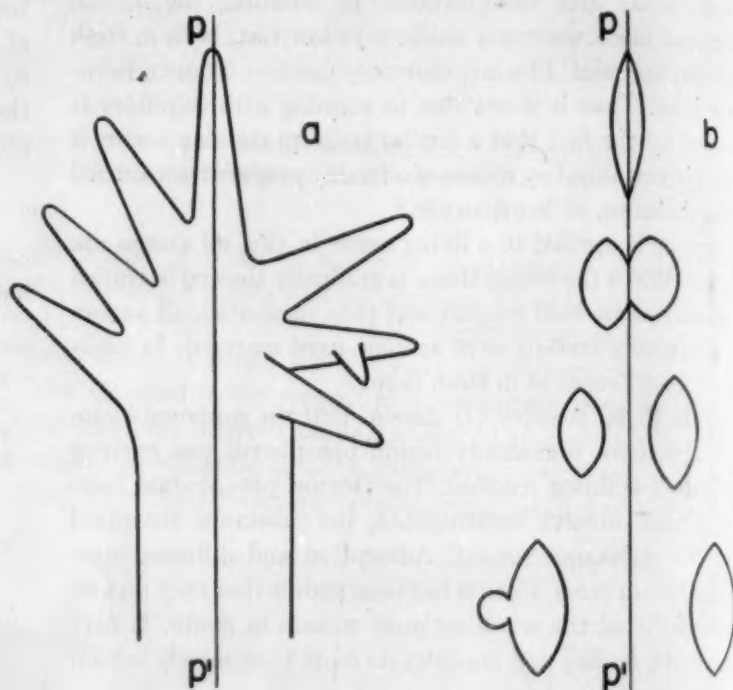


FIG. 2. Possible mirror-image symmetry in limb duplications: (a) anteroposterior symmetry in right limb of spec. #7; (b) dorsoventral symmetry in extra limb of spec. #8 (p-p' represents the plane of symmetry).

in the present series; most of the limbs show no such symmetry but rather appear to have added extra digits in series, adjacent to digit #5, as clearly shown in the right hind limb of specimen #10. (Fig. 1b).

Investigations of these polydactylous salamanders are in progress to determine the nature of the genetic background and those environmental factors, e.g. cold shock, which might have contributed to the widespread occurrence of these anomalies within a limited *Ambystoma* population.

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Antibiotic Activity of Subtilin and Streptomycin in the Presence of BAL¹

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The sulfhydryl groups in enzymes have been stressed by some authors as responsible for the attack of antibiotic (4) and metallic antiseptics (9) on microorganisms as well as for the arsenic poisoning of mammalian tissues (1). The inactivation of antibiotic substances by sulfhydryl compounds in certain cases (6, 7) and the effectiveness of using BAL as an antidote to arsenic and other heavy metal poisoning (3, 11, 14) gave support to this postulation.

BAL (2,3-dimercaptopropanol), a simple compound with two sulfhydryl groups, has been reported by Johnson, *et al.* (12) to be able to inactivate bacitracin, an antibiotic produced by a strain of *B. subtilis*. Subtilin, which is also produced by a strain of *B. subtilis*, has an antitubercular activity *in vitro* comparable to that of streptomycin. This property of subtilin is being carefully studied in this laboratory. Chin (5) has reported that sodium chloride does not alter the antibiotic action of subtilin.

synergistic with subtilin in bacteriostatic action on all strains of mycobacteria studied but not on *M. lysodeikticus*. The antibiotic action of streptomycin was affected in various ways. The bacteriostatic activity against pathogenic mycobacteria was potentiated, though not as remarkably as in the case of subtilin, whereas against the nonpathogenic strain #599 its action was not affected. Streptomycin was inactivated in the presence of BAL against *M. lysodeikticus*.

The mechanism of synergism between subtilin and BAL is entirely unknown. A similar potentiation, though to a lesser degree, of the action of streptomycin against a pathogenic mycobacterium is remarkable and may reflect that BAL itself acted somehow on the organisms so that they were rendered more susceptible to antibiotics.

Shwartzman (15) has reported synergism between methionine, methionine sulfoxide, and threonine and penicillin and suggested that these amino acids reverse the action of other antagonistic substances. The same explanation is unlikely and not applicable to the case of BAL, which potentiates both subtilin and streptomycin similarly.

BAL has been known to attack enzyme systems (17). Is it possible that some enzyme systems of mycobacteria are susceptible to BAL? Since BAL has a structure very similar to

TABLE 1
EFFECT OF BAL ON THE BACTERIOSTATIC LEVEL OF SUBTILIN AND STREPTOMYCIN

Organism	Effective level of subtilin		Effective level of streptomycin		Conc. of BAL
	with BAL	without BAL	with BAL	without BAL	
Pathogenic <i>Mycobacterium</i>					
Human H37Rv	1:32 × 10 ⁶	1:2 × 10 ⁶	1:32 × 10 ⁶	1:8 × 10 ⁶	1:10 ⁶
Human R1	1:64 × 10 ⁶	1:4 × 10 ⁶	1:64 × 10 ⁶	1:16 × 10 ⁶	1:10 ⁶
Bovine	1:128 × 10 ⁶	1:32 × 10 ⁶	1:64 × 10 ⁶	1:32 × 10 ⁶	1:10 ⁶
Nonpathogenic <i>Mycobacterium</i> #599	1:16 × 10 ⁶	1:8 × 10 ⁶	1:32 × 10 ⁶	1:32 × 10 ⁶	1:10 ⁶
<i>Micrococcus lysodeikticus</i>	1:16 × 10 ⁷	1:16 × 10 ⁷	1:2 × 10 ⁶	1:8 × 10 ⁶	1:10 ⁴

Since Loo, *et al.* (13) and Berkman, *et al.* (2) have reported that sodium chloride antagonized the antibiotic action of streptomycin, which was dissimilar to the case of subtilin, it is of interest to know whether sulfhydryl compounds which have been known to inactivate streptomycin (7, 16) affect subtilin similarly or dissimilarly. While experiments on other sulfhydryl compounds are still in progress, the study on BAL has brought about interesting results.

This study has been made upon four strains of *Mycobacterium*, two human strains (H37Rv and R1), one bovine, and one nonpathogenic fast-growing #599, and a strain of *Micrococcus lysodeikticus*.³ *M. lysodeikticus* was cultured in beef-heart infusion medium, and tests were also made in the same medium. Mycobacteria were cultured and tested in Dubos medium (8). Since BAL at a dilution of 1:10,000 has been found to inhibit the growth of mycobacteria, a dilution of 1:100,000 was used. The results, as shown in Table 1, indicate that BAL was

that of glycerol, which is utilized especially by pathogenic mycobacteria as a source of carbon (18), and it has been found to inhibit the growth of these organisms at 1:10,000, does it mean that Fildes' theory (10) of biochemical antagonism may be extended to the field of nonessential metabolites? Even if the action of BAL on a mycobacterium can thus be explained, the question of synergism remains to be elucidated. An elucidation of this mechanism is practically impossible at present when so much about the metabolic processes of mycobacteria remains unknown and is awaiting further study.

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¹ The authors are indebted to the Biochemical Division, Western Regional Research Laboratory, U. S. Department of Agriculture, Albany, California, for the supply of subtilin; to Eli Lilly & Co., Indianapolis, Indiana, for the supply of streptomycin and partial support of the work.

² Lilly Research Fellow in Pharmacology.

³ H37Rv was obtained from the standard culture depot, National Tuberculosis Association; R1, bovine, and #599 strains, from the Lilly Research Laboratories; *M. lysodeikticus*, from H. B. Woodruff.

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Inadequate Maternal Nutrition and Hydrocephalus in Infant Rats¹

LUTHER R. RICHARDSON and JEANNE DEMOTTIER

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Richardson and Hogan (1) observed hydrocephalus in infant rats as the result of feeding an inadequate diet to the mother. In this abnormality the head is dome shaped and greatly enlarged. The brain cavity is filled with serum and transmits light readily. In some cases the eyes are abnormally small and muscular incoordination usually develops if the affected rat survives long enough.

In order to demonstrate more conclusively that hydrocephalus is caused by a nutritional deficiency it seemed desirable to produce the abnormality in an unrelated colony of rats. The colony at the Texas Station, which has been maintained for more than 10 years without the introduction of any new strains, was suitable for such a study. A total of 38 females have received Diet A, which is essentially the same in composition as that used by Richardson and Hogan. This diet is composed of casein (acid washed), 25 grams; Cerelose, 57 grams; wood pulp, 3 grams; salts, 5 grams; lard, 10 grams; choline chloride,² 0.1 gram; inositol,² 0.01 gram; p-aminobenzoic acid,² 0.05 gram; vitamin A, 3,000 I.U.; vitamin D, 425 I.U.; α -tocopherol,² 2.5 mg.; Menadione,² 2.5 mg.; thiamine chloride,² 1.0 mg.; riboflavin,² 1.0 mg.; pyridoxine hydrochloride,² 1.0 mg.; calcium pantothenate,² 4.0 mg.; niacin,² 5.0 mg.; and biotin,² 0.02 mg.

Some of the experimental females were from mothers which received a stock diet and others were from mothers which received a synthetic diet, but in every case they received Diet A from 28 days of age until the observations were discontinued. A female was observed until it was evident that she would not produce any additional young.

A total of 10 young have developed typical hydrocephalus. The incidence was 1.5 per cent, or approximately the same as that given in the earlier report. None has occurred in the offspring of females which received a stock diet composed of

natural feedstuffs. The hydrocephalus in 5 of the 10 young was identified at birth. It was identified again in these same young when they were 10 days old by observing the transmission of light through the brain cavity, and finally by autopsy. It was not identified in the other 5 until they were about 10 days old. Twelve additional young appeared to be hydrocephalic at birth, but none of these survived longer than two days, and these early identifications have not been entirely reliable.

Richardson and Hogan observed one case of hydrocephalus in the offspring of a mother which received 5 per cent of dried yeast in the diet. This observation suggested that a small amount of yeast in the diet would furnish very little, if any, of the factor which prevents hydrocephalus, and at the same time it would supply sufficient pteroylglutamic acid for normal reproduction.

Intestinal synthesis of some unrecognized factor might decrease the incidence of hydrocephalus, even though the diet itself was low in this factor. The addition of a sulfonamide to the diet would decrease this intestinal synthesis and thus increase the incidence of the abnormality. In order to test this possibility 12 females were given Diet B, which is the same as Diet A with 2 per cent of dried yeast and 1 per cent of sulfasuxidine substituted for equal amounts of Cerelose. So far, these 12 females have produced 92 young and none has been hydrocephalic. These data are too insufficient to be conclusive, but they indicate that the addition of sulfasuxidine to the diet does not increase the incidence of the abnormality under these conditions.

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Effect of Flavonols on *Clostridium botulinum*

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While conducting experiments on the microbial spoilage of vegetables, the authors found that, although asparagus is readily attacked by many organisms, it is a poor medium for growth of *Clostridium botulinum*. The thought was entertained that the flavonol compound described by Campbell (1) as occurring plentifully in asparagus, and subsequently shown by DeEds and Couch (2) to be rutin, might be responsible. Following the report by Naghski, Copley, and Couch (3) on the suppression of *Staphylococcus aureus* by quercetin, an aglucone derivative of rutin, tests were made to determine the action of rutin, quercetin, and quercitrin² (a rhamnoside of quercetin) on *Cl. botulinum*. Three sets of flasks, each flask containing 15 grams of green peas and 15 ml. of corn steep-casein medium, were inoculated with a mixture of approximately 1,000 detoxified spores of *Cl. botulinum*, Types A and B, per flask. The reaction of the medium was pH 6.60. One flask in each set was left untreated as control. Weighed amounts

¹ Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture.

² Thanks are due F. DeEds, Pharmacology Laboratory, and J. F. Couch, Eastern Regional Research Laboratory, Bureau of Agricultural and Industrial Chemistry, for furnishing quercetin, quercitrin, and rutin for use in these experiments.

¹ Authorized for publication as technical article No. 1057, Texas Agricultural Experiment Station.

² Kindly supplied by Merck and Company, Rahway, New Jersey.

of rutin, quercetin, and quercitrin were added to give concentrations ranging from 60 to 160 p.p.m. The flasks were heated for 5 minutes at 100° C. and incubated anaerobically for 10 days at 23° C. Sterile filtrates were then prepared and tested for toxin by inoculating 0.20-ml. amounts into mice.

Rutin in concentrations up to 1,000 p.p.m. did not prevent growth of *Cl. botulinum* or interfere with toxin production. Quercitrin was effective only in concentrations of about 1,000 p.p.m. The action of quercetin was, however, well marked, amounts of from 80 to 160 p.p.m. in the course of several trials preventing toxicity. In one experiment, using corn steep-casein medium alone, no toxin was demonstrated in a concentration of quercetin of 20 p.p.m. The action is antibacterial, only an occasional cell appearing in smears after incubation. Concentrations of quercetin as high as 1,000 p.p.m. failed, however, to inactivate preformed toxin of *Cl. botulinum* in 72 hours.

The limited action of quercitrin in preventing growth of *Cl. botulinum* in the present experiment may have been due to the presence in the sample of a small amount of quercetin. Also, it is considered possible that in certain samples of asparagus enough quercetin may naturally be present to check growth of *Cl. botulinum*. As lately shown by Naghski, Copley, and Couch (4), the compound is antagonistic to *Brucella abortus* and *Aerobacillus polymyxa* as well as to staphylococci. Whether anaerobic organisms in addition to *Cl. botulinum* are affected is at present uncertain.

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IN THE LABORATORY

A Flower Marker for Plant-breeding Operations

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Wherever controlled cross-pollinations are made on a large scale, any simplification of the method of marking flowers is desirable. String tags, which are often used for this purpose, are tedious to apply, especially to smaller flowers; require pencil or ink marking; and are susceptible to weather damage, which may even obliterate identification markings.

The requirements for this type of marker have been met economically by small pieces of "Twist-Ems"—a product devised for tying plants and for bunching root vegetables, consisting of a wire strand sealed by a waterproof adhesive between two narrow strips of heavy paper. These markers, used here for periods of three to five months, have proved to be quite weather resistant; moreover, they have been so durable that it has been possible to salvage them for satisfactory use in a second season.

When used to tag pollinated tomato flowers, "Twist-Ems" are cut into inch lengths. Each piece is folded at one end, placed on the flower so that the pedicel lies within the fold, and then folded in the same direction at the other end (Fig. 1). The second fold serves to lock the first one and thus prevents loss of the marker. Any identification markings can be protected from weathering by keeping them inside the folds. Although firmly attached, the marker will not prevent further growth; the wire of "Twist-Ems" is so easily bent that it does not constrict the developing pedicel.

Markers intended for various crosses can be distinguished by painting them different colors in fade-proof lacquers. Two or more colors can be applied in various combinations to the

same marker in longitudinal or oblique stripes, thereby increasing greatly the number of different identifiable markers. Various combinations of letters or numbers (dates if desired) can be printed on the strips by means of an improvised rotary rubber stamp. The parents of a particular cross combination

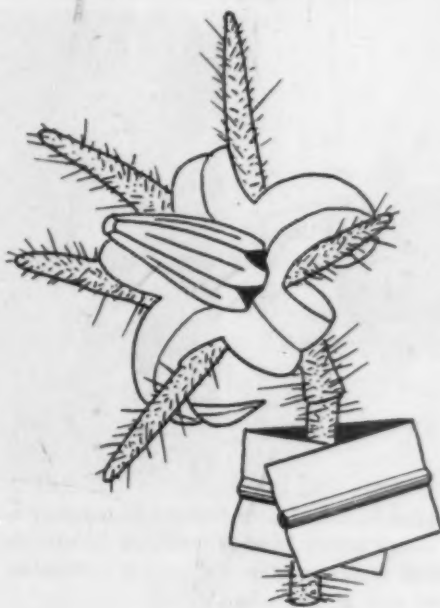


FIG. 1. Diagram of the flower marker placed on the pedicel of a tomato flower.

can then be identified by reference to a key to the color or number combinations.

These markers have been used successfully for tagging flowers of tomatoes, asparagus, and cabbage as well as in marking stems and petioles of particular ages where later identification was needed. Many other uses in biological research might be found where large-scale marking is required.

Mounting of Dermatological Material With Synthetic Resins

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In order to have available, especially for teaching and investigative purposes, permanent mounts of cleared, unstained, fungus-infected materials, trials have been made with the various mounting materials currently available. Colorless nail polish has also been used. To date, one of the most satisfactory of these mounting materials has been the Permout (obtainable from Fisher Scientific Company, Pittsburgh). After this report was prepared, the recent work of Barghoorn (1) on phenol formaldehyde and vinyl resins was found. Permout

tans, *Phthirus pubis* with ova, *Pediculus capitis* and *corporis* and *Cimex lectularius*. For the preservation of larger specimens, the technics of Sando (3), Strumia and Hershey (4), and Kampmeier and Haviland (2) are preferred.

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Polytetrafluoroethylene for the Prevention of "Bumping" From Superheating of Boiling Fluids

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Polytetrafluoroethylene, recently developed as a commercial plastic under the trade name of "Teflon,"¹ is extremely inert chemically, being unattacked by hot or cold acids or alkalis, or by chemical oxidizing agents; appears to be insoluble in all aqueous, organic, and silicone fluids; is attacked by molten sodium or potassium; is not wet by aqueous fluids or by many organic fluids; and can be cleaned in strong acid cleaning solutions or in alkali.

A sample piece of "Teflon" sheeting 0.040 inch thick was kindly supplied by the DuPont Company over a year ago. While cleaning some of the material in hot, 30 per cent sodium hydroxide solution it was noted that the solution boiled smoothly for many hours without bumping. Since then, small bits of polytetrafluoroethylene have been used routinely to prevent bumping in boiling fluids. The material has worked with uniform success with alkaline and acid solutions except as noted below, and with numerous organic fluids, including aniline, propionic acid, decahydronaphthalene, morpholine, ethylene dichloride, chloroform, benzyl alcohol, amyl acetate, ethyl alcohol, acetone, and many others.

It has not been very satisfactory as an antibumping agent for Kjeldahl digestions, probably because it floats in this heavy acid mixture.

It is striking that polytetrafluoroethylene retains its antibumping action for many hours—probably indefinitely. Pieces can be used repeatedly without evidence of deterioration.

One is tempted to theorize that antibumping action is probably related closely to nonwetability. Perhaps only an immersed surface not wet by the fluid brings about a satisfactory liquid boundary from which vapor molecules can escape readily. This might explain the well-known fact that glass beads function as satisfactory antibumpers when freshly added to an alkaline solution, when perhaps they are not perfectly clean and hence may have surfaces not wet well by water, whereas after a short time they cease functioning efficiently, presumably after they have been cleaned off by the solution and rendered wettable. Polytetrafluoroethylene, however, retains its nonwetting properties when perfectly clean.

¹ Manufactured by E. I. duPont de Nemours & Company.

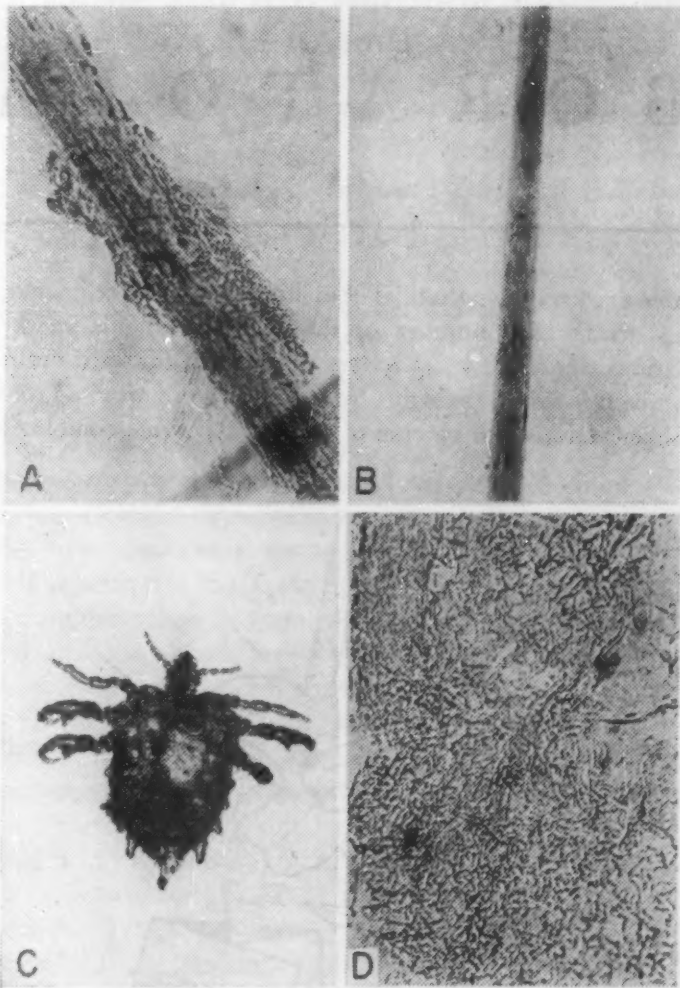


FIG. 1. Some specimens in a dermatological laboratory mounted directly in Permout: A.—hair infected with *M. audouini* (X 700); B.—hair acquired canities at zone of depigmentation (X 160); C.—*Phthirus pubis* (X 35); D.—skin infected with *M. audouini* (X 700).

has been reported merely as a 60 per cent solution of pinone polymers. Without any preparation, the hair or scales of skin are put on a slide covered with a thin coating of the solution of the resins, covered with a cover slip pressed to remove bubbles of air. The solvent may serve to clear the tissue; the plastic, to preserve it. The specimens are available for immediate examination (Fig. 1). Over a period of a year the specimens have shown no discoloration or distortion. The solution has been used also to mount satisfactorily *Simulium*, *Pulex irri-*

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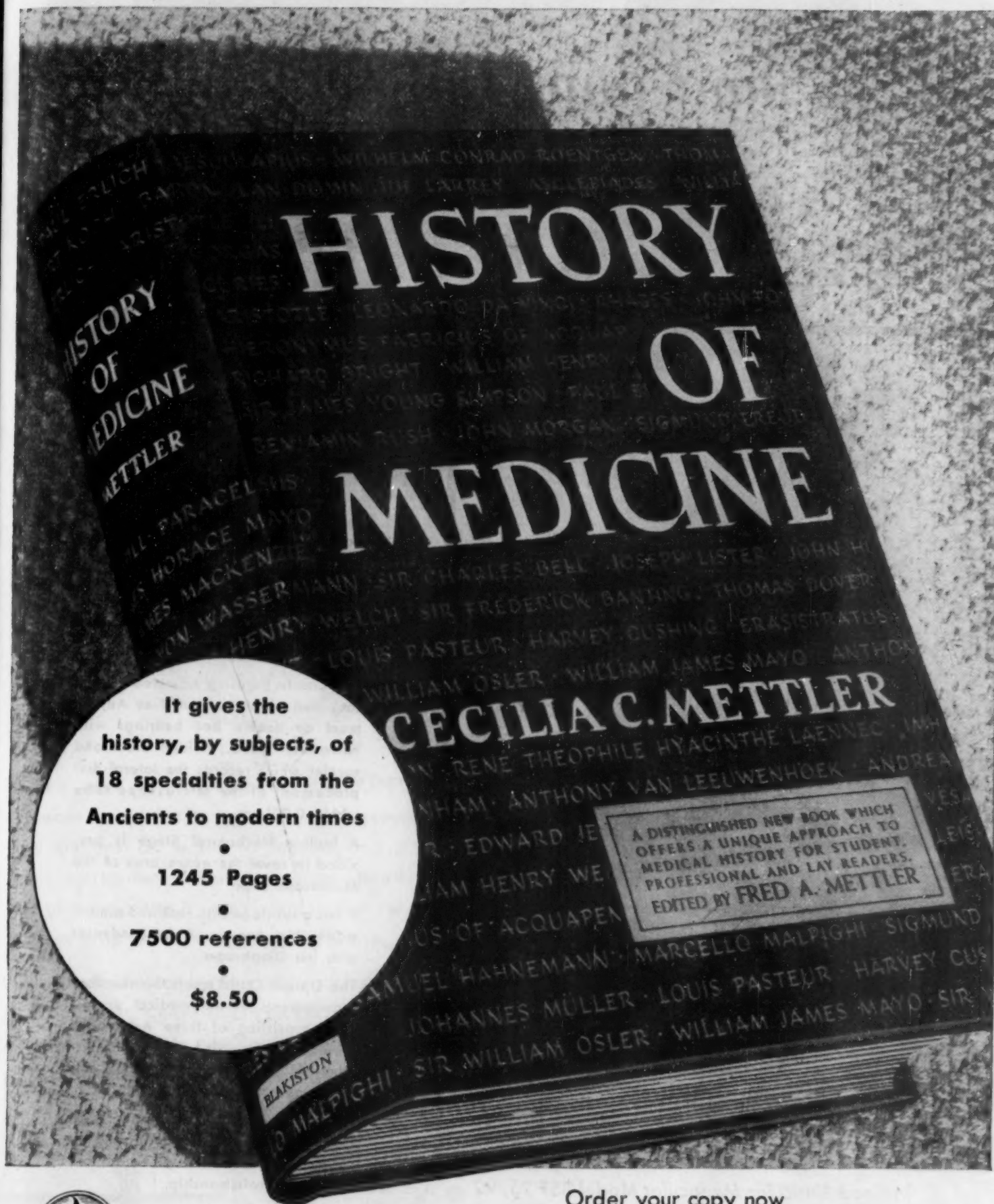
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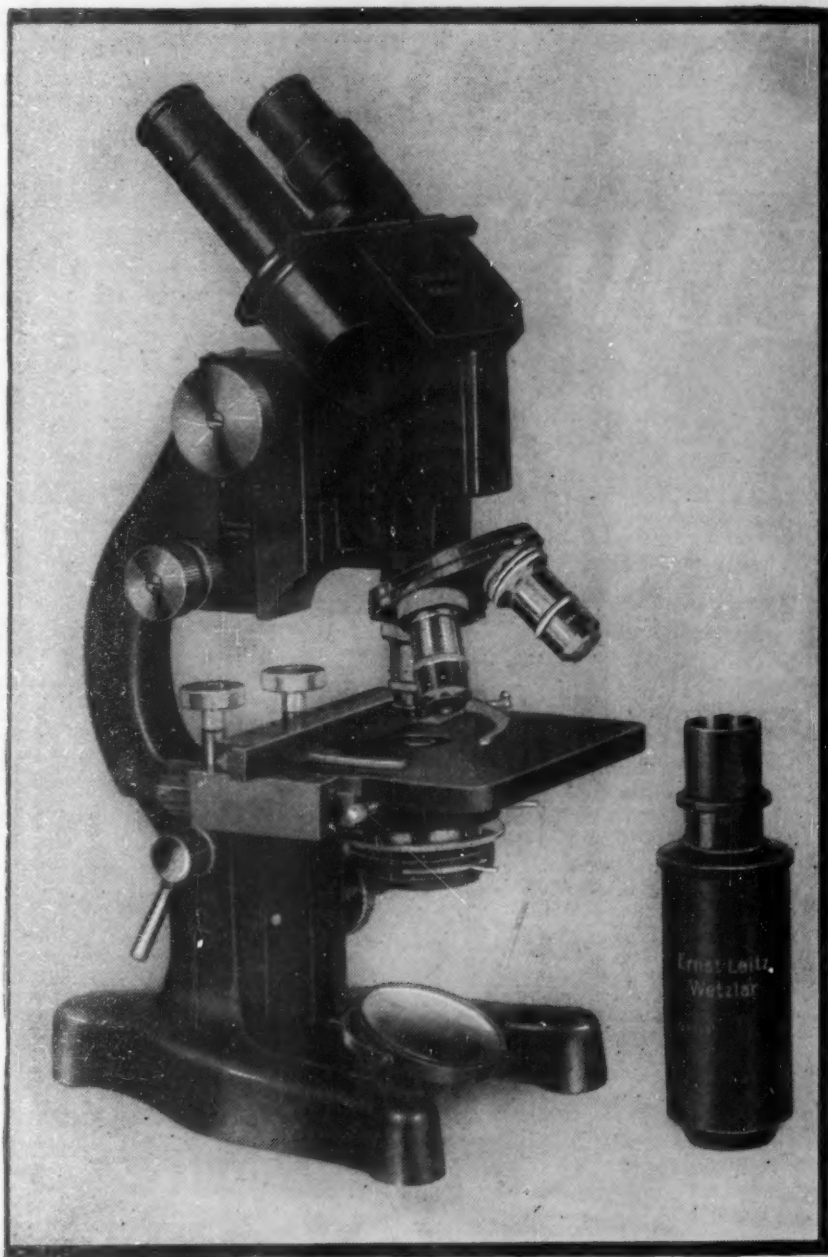
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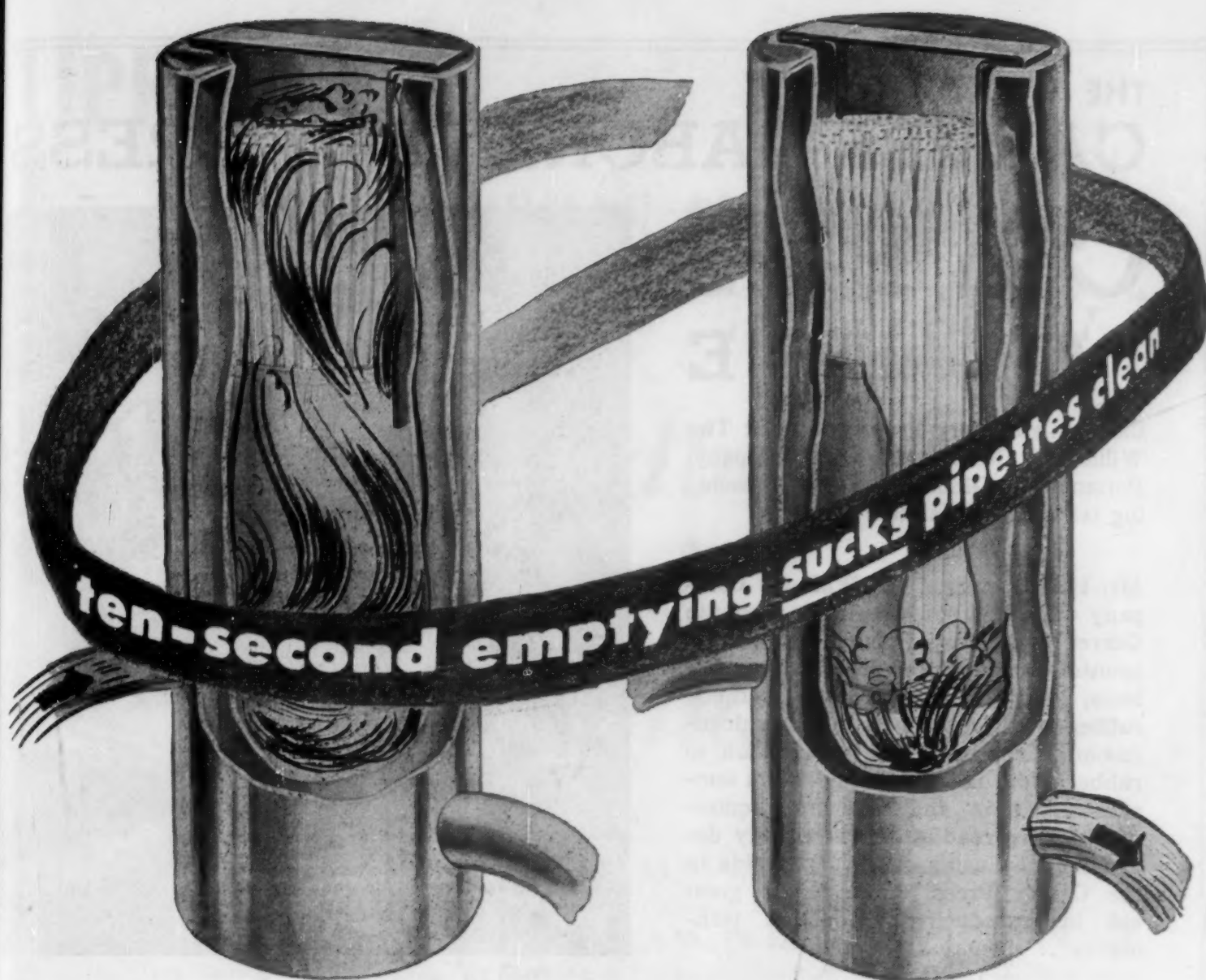
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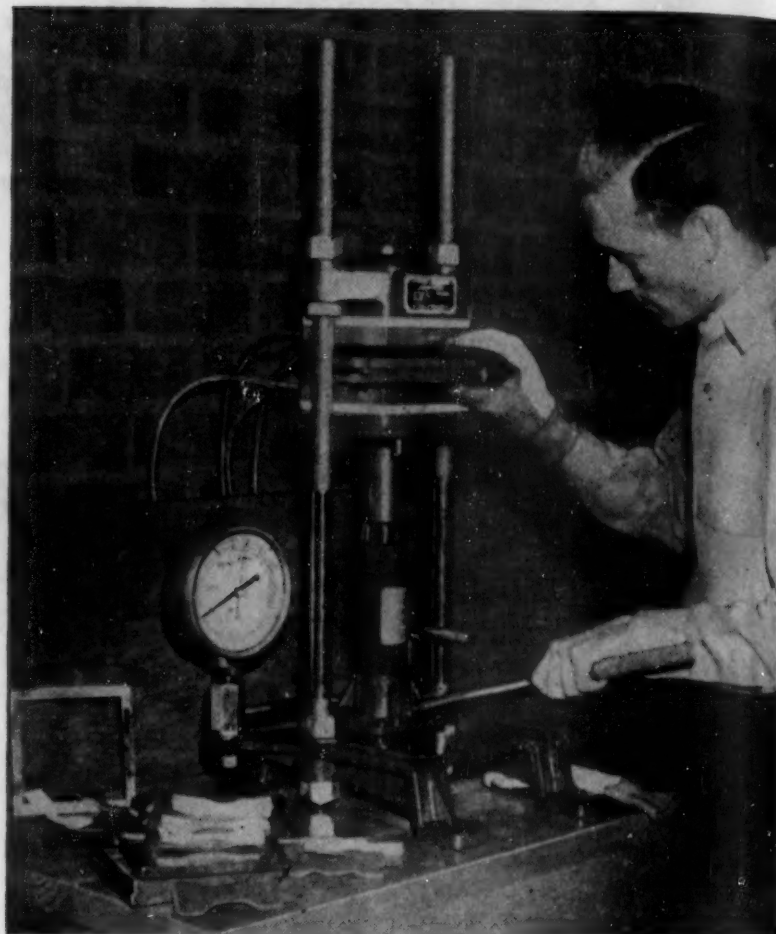


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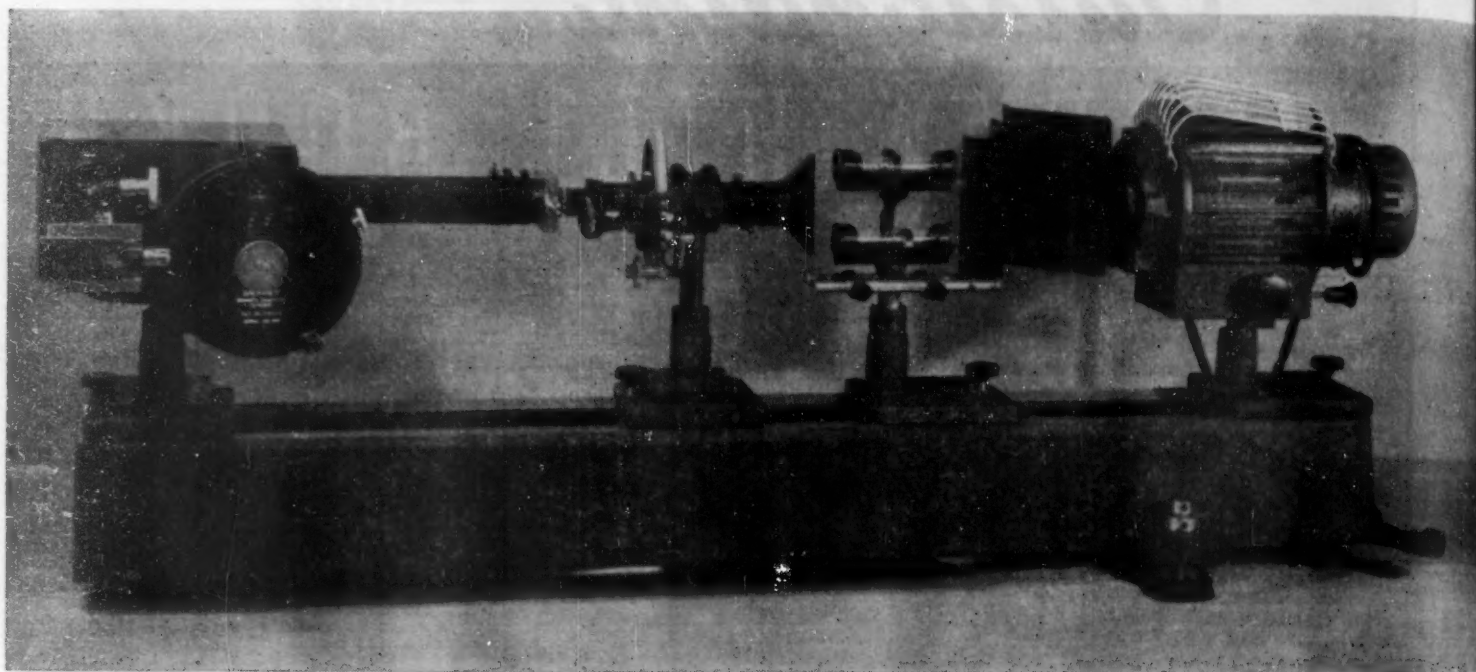
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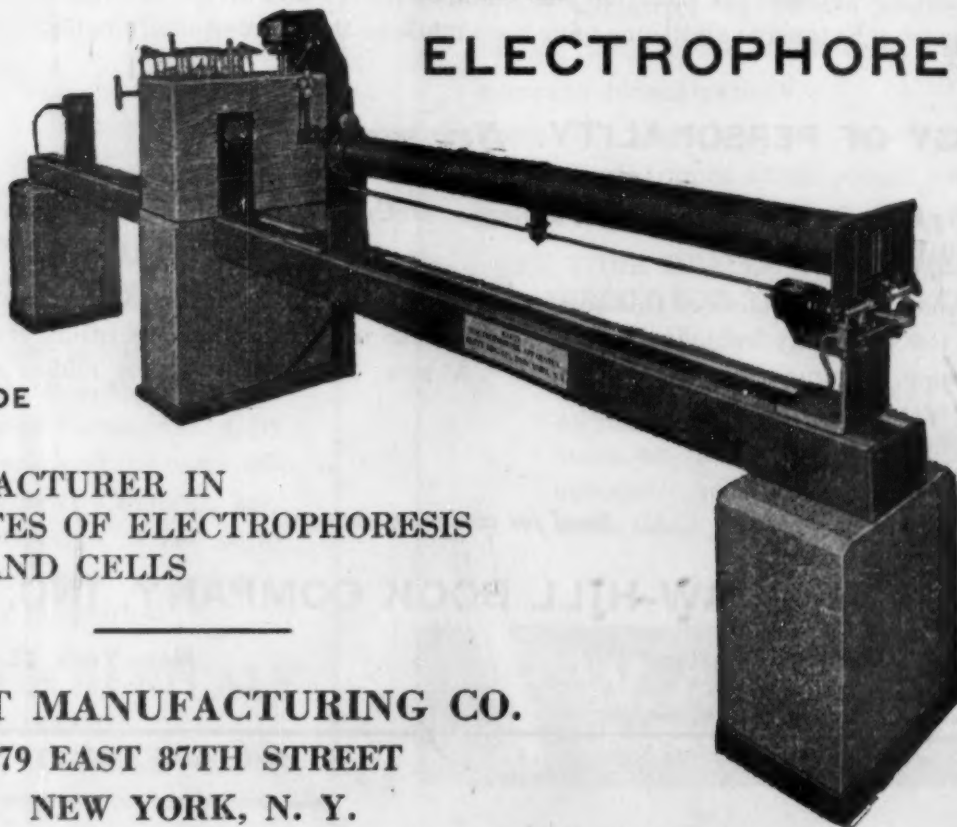
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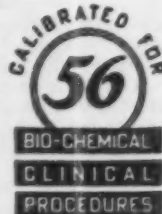
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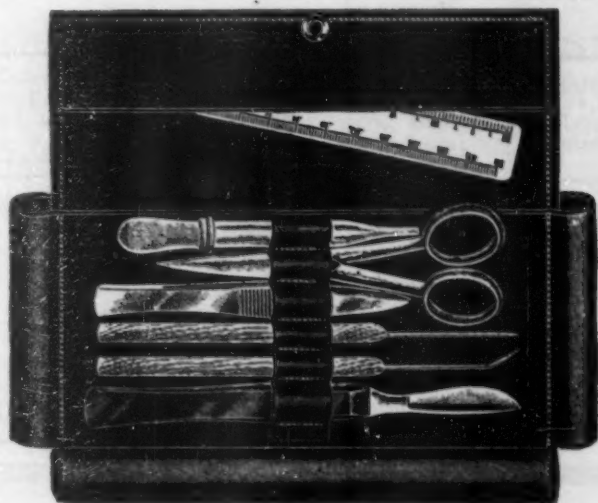
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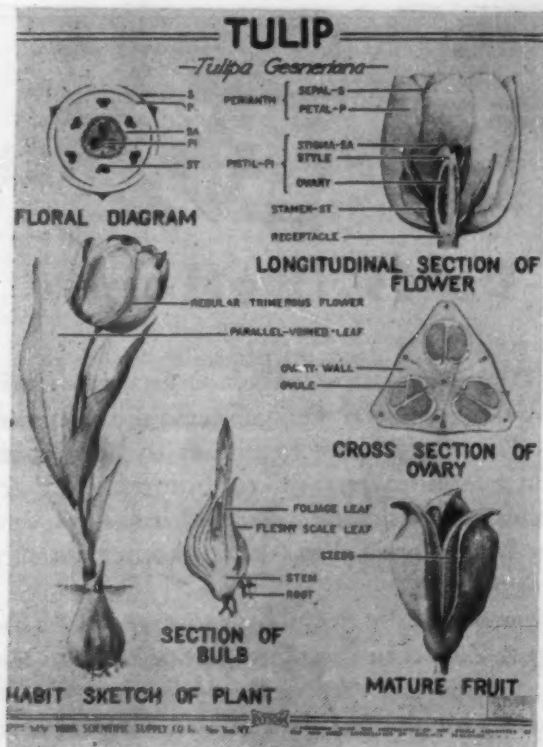
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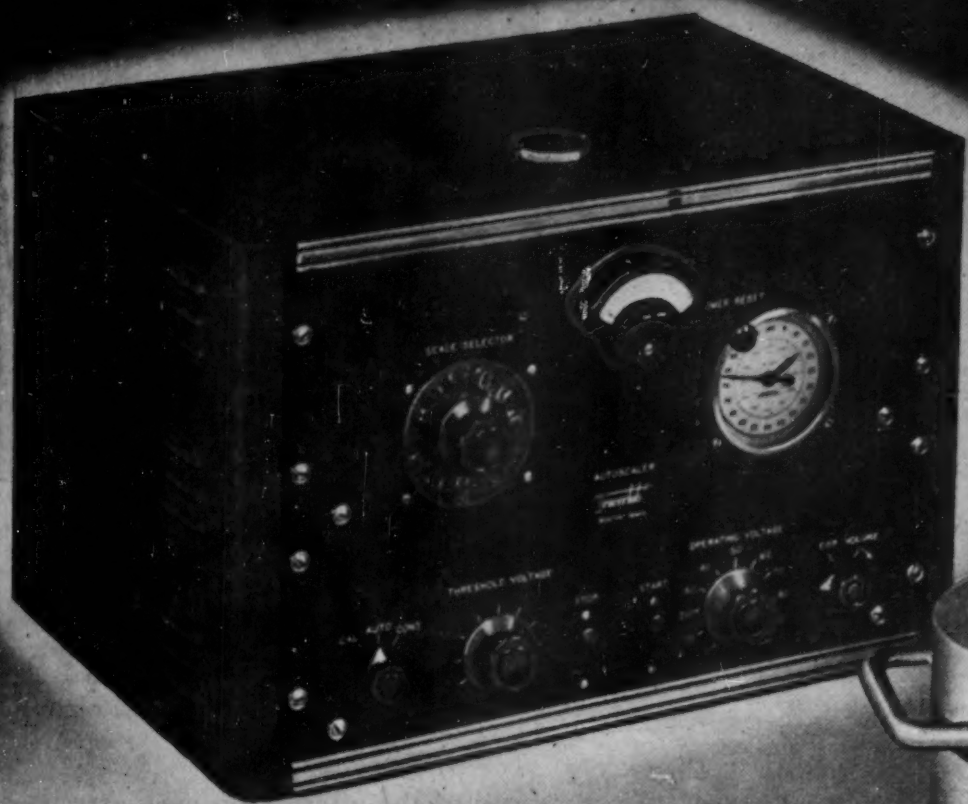
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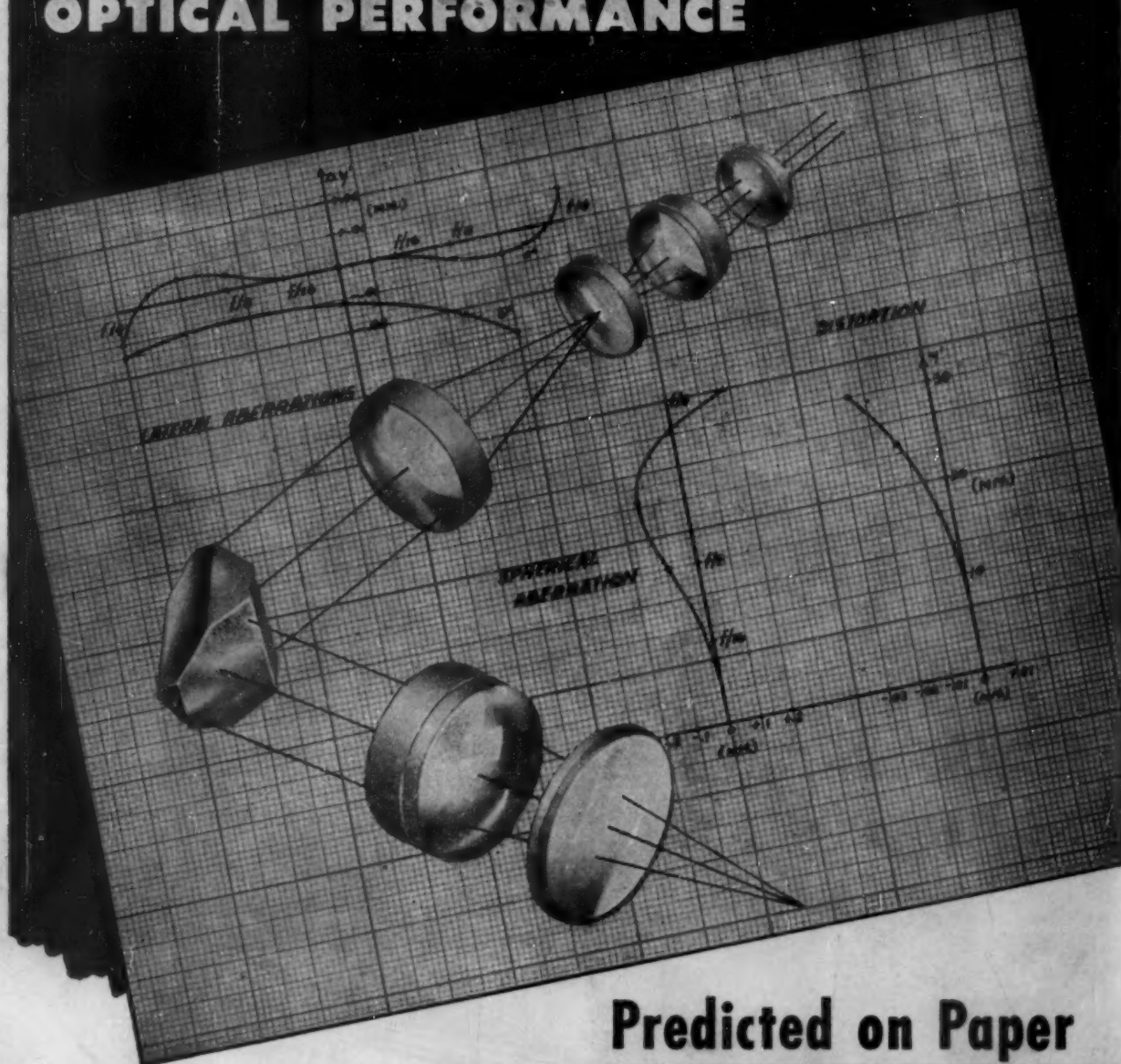


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